When citing an abstract from the 2017 annual meeting please use the format below.

[Authors]. [Abstract Title]. Program No. XXX.XX. 2017 Neuroscience Meeting Planner. Washington, DC: Society for Neuroscience, 2017. Online.

Poster

366. Fate Specification and Development of Glia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 366.01/A1

Topic: A.01. Neurogenesis and Gliogenesis

Support: Shriners Hospitals Research Fellowship 84306

Shriners Hospitals Grant 85400

Shriners Hospitals Grant 85800

NIH Grant NS025044

Title: Differing cell-intrinsic properties between forebrain and spinal cord oligodendroglial lineage cells

Authors: *M. HORIUCHI[1], Y. S. HORIUCHI[2], T. AKIYAMA[3], A. ITOH[4], D. E. PLEASURE[5], E. E. CARSTENS[6], T. ITOH[7]
[1]Lewis Katz Sch. of Med. at Temple Univ., Philadelphia, PA; [2]Dermatol., Univ. of Pennsylvania, Sch. of Med., Philadelphia, PA; [3]Dermatol., Univ. of Miami, Miami, FL; [4]Neurol., Univ. of California, Davis, Sacramento, CA; [5]Neurol, Univ. of California Davis Dept. of Neurol., Davis, CA; [6]Neurobiology, Physiol. & Behavior, Univ. of California Davis, Davis, CA; [7]Dept Neurol., Univ. California Davis, Sacramento, CA

Abstract: Differentiation of oligodendroglial progenitor cells (OPCs) into myelinating oligodendrocytes is regulated by the niche where they differentiate. However, it has not been clarified whether or not oligodendroglial lineage cells (OLCs) derived from different anatomical regions of the central nervous system (CNS) respond to microenvironmental cues in the same manner. Here, we compared pure OPCs from rat neonatal forebrain and spinal cord in the same *in vitro* conditions. We found that forebrain and spinal OLCs respond differently to the same external factors. They were distinct in proliferation responses to mitogens, oligodendrocyte phenotype after differentiation, and cytotoxic responses to α-amino-3-hydroxy-5-methyl-4-isoxazolepropionate-type glutamate receptor-mediated excitotoxicity at immature stages of differentiation in a cell-intrinsic manner. Moreover, transcriptome analysis identified genes differentially expressed between these OPC populations, including those encoding transcription factors, cell surface molecules, and signaling molecules. Particularly, forebrain and spinal OPCs retained the expression of region-specific transcription factors, such as Foxg1 and Hoxc8, respectively, even after serial passaging *in vitro* indicating that OPCs maintained regional identity of their origins through the multiple cell divisions. Given the essential role of these transcription factors in the regional identities of CNS cells along the rostrocaudal axis, our

results suggest that CNS region-specific gene regulation by these transcription factors may cause cell-intrinsic differences in cellular responses between forebrain and spinal OLCs to extracellular molecules. Further understanding of the regional differences among OPC populations will help to improve treatments for demyelination in different CNS regions and to facilitate the development of stem cell-derived OPCs for cell transplantation therapies for demyelination.

Disclosures: M. Horiuchi: None. **Y.S. Horiuchi:** None. **T. Akiyama:** None. **A. Itoh:** None. **D.E. Pleasure:** None. **E.E. Carstens:** None. **T. Itoh:** None.

Poster

366. Fate Specification and Development of Glia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 366.02/A2

Topic: A.01. Neurogenesis and Gliogenesis

Title: Prospective identification and regulation of Glast positive cells in the mouse hypothalamus

Authors: L. TIROU[1], M. DAYNAC[1], H. FAURE[1], M.-A. MOUTHON[2], F. BOUSSIN[2], *M. RUAT[1]
[1]NeuroPSI, UMR9197, CNRS, Gif Sur Yvette, France; [2]CEA DSV iRCM SCSR, Fontenay aux Roses, France

Abstract: The hypothalamus (HPT), a central regulator of energy homeostasis, controls food intake, metabolism and body temperature. Emerging evidence indicates that active neurogenesis occurs in the post-natal and adult mammalian HPT and that new neurons are added to the appetite and energy-balance regulating centers in young and old animals (Sousa-Ferreira et al, *Trends Endocrinol Metab*, 2014). Tanycytes, ependymocytes, subventricular astrocytes and parenchymal glial cells that are located next to the third ventricle (3V) represent potential stem and progenitor cell candidates. However, the precise location and identity of these cells are not known yet. We have developed a conditional mouse (GlastCre^{ERT2}-YFP) reporter line where YFP is expressed in Glast^{+} cells (astrocyte-specific glutamate transporter expressed in astrocytes and astrocyte-like Neural Stem Cells (NSCs)) upon administration of tamoxifen (Daynac et al, *Stem Cell Reports*, 2016). We have observed that ten days after tamoxifen administration in young adult mice, the reporter activity is detected in all tanycyte populations at the level of the median eminence. These cells were located in the ventricular zone of the 3V and extended basal processes as shown by intense YFP signals. The signal was also evident in the parenchyma in agreement with its expression in Glast^{+} astrocytes. Using this reporter mouse model, we have purified Glast-YFP cells from HPT of adult mice by fluorescence-activated cell sorting (FACS). We labelled the YFP cells with a Glast antibody to distinguish Glast-expressing recombined cells (Glast^{+}YFP^{+}) from their progeny that no longer express Glast (Glast^{-}YFP^{+}). Experiments are in progress to further quantify and characterize YFP expression in Glast^{+} cells and in their progeny

6 and 10 months after tamoxifen administration and to delineate the molecular mechanisms (single cell RNAseq) regulating the maintenance of these cells. These experiments will bring novel methods and genetic tools to investigate the role of Glast$^+$ cells and their progeny in feeding behaviors, energy-balance and in neurodegenerative diseases.

Disclosures: L. Tirou: None. **M. Daynac:** None. **H. Faure:** None. **M. Mouthon:** None. **F. Boussin:** None. **M. Ruat:** None.

Poster

366. Fate Specification and Development of Glia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 366.03/A3

Topic: A.01. Neurogenesis and Gliogenesis

Title: Pdgfra$^+$ progenitor cells differentiate in both oligodendrocytes and pericytes in the developing mammalian central nervous system

Authors: *E. M. FLORIDDIA, D. DIAS, S. MARQUES, C. GÖRITZ, G. CASTELO-BRANCO
Karolinska Institutet, Stockholm, Sweden

Abstract: Oligodendrocytes (OLs) and pericytes (PCs) have very distinct functions. OLs myelinate and metabolically support axons of the central nervous system, while PCs are involved in angiogenesis, blood flow regulation, and are essential to maintain the integrity of blood-brain barrier. We have recently identified a *Pdgfrα*$^+$ cell subpopulation (a widely accepted marker of OL precursor cells) that belongs to the pericyte lineage by single-cell RNA-sequencing analysis of the OL lineage (Marques and Zeisel et al, 2016, Science). OL progenitor cells and PCs also share other markers such as NG2 (*Cspg4*). Moreover, Sox10 is a pan marker for the oligodendrocyte lineage but Sox10$^+$ neural crest progenitors have been reported to originate PCs (Simon et al, 2012, Genesis). To address whether OLs and PCs have a common multipotent progenitor, we fate mapped Pdgfrα$^+$ cells at embryonic (E) day 13.5 and observed that they differentiate in both OLs (expressing Pdgfrα and CC1) and PCs (expressing Col1α1) in the brain and spinal cord. We also fate-mapped pericytes at embryonic and juvenile stages, using the *Col1α1CreER^{T2}-TdTomato* reporter mouse line. We observed that a subpopulation of Col1α1$^+$ pericytes express markers such as Pdgfrα$^+$ and Sox10$^+$ in vitro and in vivo, but not markers of OL differentiation, such as MOG, CNPase, or MBP. We are further investigating the fate of the Col1α1$^+$ pericytes in case of demyelination. We aim to unveil whether a cell type within the pericyte lineage has the ability to transdifferentiate into oligodendrocyte and contribute to self-repair following demyelinating injuries.

Disclosures: **E.M. Floriddia:** None. **D. Dias:** None. **S. Marques:** None. **C. Göritz:** None. **G. Castelo-Branco:** None.

Poster

366. Fate Specification and Development of Glia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 366.04/A4

Topic: A.01. Neurogenesis and Gliogenesis

Support: 1R01NS096100-01

1R21MH110724-01

CURE, PA Department of Health

Title: Postnatal development of Gli1 astrocytes in the mammalian cortex

Authors: ***E. C. GINGRICH**[1], B. TING[1], O. OLUKOYA[3], M. WALOGORSKY[3], C. HARWELL[3], A. D. R. GARCIA[2]
[2]Neurobio. and Anat., [1]Drexel Univ., Philadelphia, PA; [3]Neurobio., Harvard Univ., Boston, MA

Abstract: Astrocytes encompass a large and heterogeneous population of cells whose molecular and functional diversity remain poorly understood. We previously showed that in the adult mammalian cortex, a subpopulation of astrocytes express the transcription factor, Gli1, indicating active and high level Sonic hedgehog (Shh) signaling. These Gli1 astrocytes exhibit a layer specific distribution such that Layers 4 and 5 exhibit the highest density of Gli1 astrocytes, while Layers 1 and 2/3 are relatively sparse. The developmental origins of cortical Gli1 astrocytes are not known. In addition, whether Shh signaling in astrocytes is restricted to mature, post-mitotic cells or whether it occurs in astrocyte precursors that produce Gli1 cells remains to be determined. In this study, we examined the developmental origins of cortical Gli1 astrocytes across postnatal development. We found that the number of cells expressing Gli1 increases dramatically over the first two postnatal weeks, reaching adult expression levels by postnatal day 14 (P14). Proliferation studies with BrdU showed that few Gli1 cells are dividing at P7, although 30% of cells are double labeled at P3, suggesting that the early postnatal cortex harbors a population of Gli1 expressing progenitor cells. In order to map and trace the lineage of proliferating, Gli1 expressing precursors, we performed genetic inducible fate mapping (GIFM) using Gli1CreER mice crossed with the Ai14 Rosa26 tomato reporter line (*Gli1*$^{CreER/+}$*;R26*$^{tdTom/tdTom}$). In animals that received tamoxifen at P7 and were analyzed at P14, marked cells show a distribution consistent with that observed in adult tissues, with the majority of cells localized in Layer 5. In contrast, animals that received tamoxifen at P3 show numerous marked cells in superficial cortical layers. Additionally, we observed a greater number of marked

cells at P3, compared with P7, suggesting that proliferation of Gli1 cells occurs primarily during early postnatal development. Single cell, colocalization analysis with tomato and cell-type specific markers showed that the vast majority of Gli1 cells marked by tamoxifen at any age express S100ß, suggesting that these cells predominantly generate astrocytes. We did not observe any marked cells that correspond to oligodendrocytes. Taken together, these data suggest that Gli1 astrocytes are derived from a Gli1 expressing precursor cell in the early postnatal cortex. Moreover, these data show that in contrast to the embryonic CNS, where Shh regulates specification and proliferation of oligodendrocyte precursor cells, Shh signaling in the early postnatal cortex occurs predominantly in astrocyte progenitor cells.

Disclosures: E.C. Gingrich: None. **B. Ting:** None. **O. Olukoya:** None. **M. Walogorsky:** None. **C. Harwell:** None. **A.D.R. Garcia:** None.

Poster

366. Fate Specification and Development of Glia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 366.05/A5

Topic: A.01. Neurogenesis and Gliogenesis

Title: FGF signaling directs neural stem cells in the subventricular zone toward oligodendrocyte lineage and improve cell regeneration after demyelination

Authors: *W. KANG[1], J. M. HEBERT, 10461[2]
[1]Albert Einstein Col. of Med., Bronx, NY; [2]Dept. of Neurosci., Albert Einstein Col. of Med., Bronx, NE

Abstract: FGF signaling plays a critical context-dependent role in both dorsal-ventral cell fate specification and neurogenesis during brain development. But its role in the regulation of adult stem/progenitor cells in the subventricular zone (SVZ) in vivo is not clear. Here in this study, a conditional mouse genetic approach was used to modulate FGF signaling specifically in adult neural stem/progenitor cells in the SVZ, and the effects of both loss and activation of FGF signaling on stem/progenitor cell fate specification was examined. The results showed that although FGF signaling is not essential for the neurogenesis in the SVZ, its increased activity leads to a greatly increased generation of Olig2+ transit amplifying progenitor (type C) cells that are committed to oligodendrocyte lineage at the expense of neurogenic progenitor cells. Importantly, upon demyelination, the increased generation of oligodendrocyte lineage progenitor cells in the SVZ results in an improved cell regeneration and replacement in the corpus callosum, whereas the neurogenesis from the SVZ is temporarily decreased, likely due to the re-directed differentiation of stem/progenitor cells toward oligodendrocyte lineage. Whether the oligodendrocyte fate specification by FGF signaling can lead to improved remyelination and functional recovery after demyelination will be further examined. The finding that the fate of

neural stem cells in the SVZ can be re-specified toward oligodendrocyte fate by FGF signaling has important implications for the potential use of endogenous stem/progenitor cells in demyelinating diseases.

Disclosures: W. Kang: None. **J.M. Hebert:** None.

Poster

366. Fate Specification and Development of Glia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 366.06/A6

Topic: A.01. Neurogenesis and Gliogenesis

Support: National Multiple Sclerosis Society

Title: Deletion of integrin linked kinase in neural progenitor cells disturbs neuron/glia balance during early spinal cord development

Authors: *R. HUSSAIN[1,2], W. B. MACKLIN[3]
[1]Dept. of Cell and Developmental Biol., Univ. of Colorado Sch. of Med., Aurora, CO; [2]Dept. of Neurosurg., Univ. of Rochester, Center for Translational Neuromedicine, NY; [3]Dept Cell & Dev Biol., Univ. Colorado Med. Sch., Aurora, CO

Abstract: Integrin linked kinase (ILK) is a serine/threonine protein kinase, localized at the inner margin of the plasma membrane, where it also serves as a scaffolding protein/focal adhesion mediator. Its presence has been reported in neurons, astrocytes and oligodendrocytes in the central nervous system, but its exact role during the embryonic development of the glial/neuronal profile in brain and spinal cord remain unknown. We selectively knocked out ILK in embryonic neural progenitor cell populations in Cre-lox experiments driven by promoter of the the 2'3 cyclic nucleotide phosphodiesterase (CNP) gene or the promoter of the transcription factor Olig1 gene. These promoters are both active in spinal cord by E12.5. Deletion of ILK severely reduced the number of proliferating (Ki67+) oligodendrocyte progenitor cells (OPCs) in both transgenic lines, which ultimately resulted in reduced OPCs in different regions of the embryonic spinal cord. ILK deletion not only reduced OPC proliferation, but also increased the number of apoptotic OPCs, identified by TUNEL staining. $CNP^{cre+/-}$ X $ILK^{fl/fl}$ (CNP-ILK cKO) embryos were embryonic lethal, and even $Olig1^{cre+/-}$ X $ILK^{fl/fl}$ (Olig1-ILK cKO) embryos often died in utero. Importantly, Olig1-ILK cKO embryos had altered neuron/glia ratios by E14.5. As detected by NeuN, Pax6, GABA, ASCL1, ISLET1, and HuD immunostaining, in the absence of ILK, the normal neuron-glia balance shifted towards production of more neuronal cells. In conclusion, ILK is important for the normal neuron-glia profile during development of the embryonic spinal cord, and its absence reduces oligodendrocyte number by reducing proliferation and increasing apoptosis.

Disclosures: **R. Hussain:** None. **W.B. Macklin:** None.

Poster

366. Fate Specification and Development of Glia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 366.07/A7

Topic: A.01. Neurogenesis and Gliogenesis

Support: NMSU Manasse Endowment

Title: Impact of growth factors on human neuroglial cell morphology

Authors: ***V. B. KNIGHT**, T. M. NUNN, E. E. SERRANO
Biol., New Mexico State Univ., Las Cruces, NM

Abstract: Understanding the plasticity of human neural cell types and neural cell fate decisions are critical milestones in the path toward the regeneration of nervous tissue after injury. A primary normal human astrocyte (NHA) cell line with phenotypic plasticity was chosen for the evaluation of the potential role of growth factors in cell fate decisions. Using phase contrast microscopy, we made a morphological assessment of NHA cell fate after 10 days in culture under spontaneous (Knockout DMEM/F-12, Glutamax, and StemPro Neural Supplement; Gibco) and neuronal (Neurobasal Media, Glutamax, and B27 Supplement; Gibco) differentiation conditions. Metamorph® software was used to evaluate (1) the number of phase-bright structures, and (2) the length of processes protruding from the cell body in NHA cultured in the two distinct growth factor combinations. Objects with a width less than 180 nm and a length more than 900 nm were considered processes. In some cases, a phasedark irregularly shaped structure interrupted objects otherwise considered processes. Objects were nevertheless counted as processes, and the number of phasedark irregularly shaped structures was tallied for each image. Our findings suggest that both the number of long processes and the number of phase-bright structures are increased in NHA cultured under neuronal differentiation conditions. Additional studies will compare neuronal beta tubulin in NHA cultured with the distinct growth factor combinations. Research supported by the NMSU Manasse Endowment.

Disclosures: **V.B. Knight:** None. **T.M. Nunn:** None. **E.E. Serrano:** None.

Poster

366. Fate Specification and Development of Glia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 366.08/A8

Topic: A.01. Neurogenesis and Gliogenesis

Title: Generation of cortical oligodendrocytes from dorsal forebrain radial glia requires Shh signaling during embryonic development

Authors: *C. WINKLER, S. FRANCO
Univ. of Colorado Sch. of Med., Aurora, CO

Abstract: Radial glial cells (RGCs) in the ventral and dorsal forebrain can give rise to committed oligodendrocyte precursor cells (OPCs) that migrate into the cortex in successive waves to generate mature oligodendrocytes. The vast majority of oligodendrocytes in the mature cortex arise from the dorsal Emx1 domain, although it has previously been thought that dorsally-derived OPCs only begin to arise during postnatal ages in the mouse forebrain. We now show that dorsal RGCs in the embryonic mouse forebrain begin to produce large numbers of OPCs as early as E15.5, during the peak of upper layer neurogenesis. Furthermore, we have identified a subpopulation of dorsal RGCs, marked by expression of the transcription factor Ascl1, that generate only cortical oligodendrocytes, but not excitatory projection neurons or astrocytes. We also show that the sonic hedgehog (Shh) signaling pathway is critical for generating these dorsal $Ascl1^+$ RGCs and for producing normal numbers of cortical OPCs and oligodendrocytes. Using a variety of Cre mouse lines to knock out Shh from various regions of the embryonic forebrain, we provide evidence that the embryonic cerebral spinal fluid is a critical source of Shh for generating oligodendrocytes from dorsal RGCs. Taken together, our results suggest a model in which a late embryonic Shh signal initiates a neuron-glia switch in a subset of dorsal RGCs to drive cortical oligodendrogenesis.

Disclosures: C. Winkler: None. **S. Franco:** None.

Poster

366. Fate Specification and Development of Glia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 366.09/A9

Topic: A.01. Neurogenesis and Gliogenesis

Title: Barx2 can drive expression of astrocytic and neuronal markers similar to glioma "asteron" cells

Authors: C. RUSSO[1], K. MCKAY[2], *M. K. TAYLOR[3]
[1]LabMate, Boston, MA; [2]Central Michigan Univ., Mt. Pleasant, MI; [3]Biomed. Sci., Grand Valley State Univ., Allendale, MI

Abstract: The characterization of candidate regulators of stem cell differentiation can be performed in the chick embryo using quantitative PCR and anatomical techniques. Here we focus on one gene of interest, Barx2, on neural progenitor differentiation in the developing chick neural tube. Barx2 is highly enriched in neural stem cell populations, making it an interesting target of study in terms of stem cell self renewal. However, overexpression of Barx2 resulted in the formation of hybrid cells that ectopically express both neuronal and astroctyic markers. These features resemble “asterons”, previously reported both as intermediate cells in neurosphere transdifferentiation in vitro and as prevalent cell types in many forms of aggressive glioma tumors. Additionally, we found that Barx2 drove the expression of multiple proteins known to be highly expressed in astrocytoma. Future work will determine potential expression patterns of Barx2 in glioma tumors and further classify the progression of the “asteron” cell type following Barx2 overexpression.

Disclosures: C. Russo: None. **K. McKay:** None. **M.K. Taylor:** None.

Poster

366. Fate Specification and Development of Glia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 366.10/A10

Topic: B.12. Glial Mechanisms

Title: Investigating the role of astrocytes in BOLD fMRI signal

Authors: *Z. SKACHOKOVA, F. SCHLEGEL, A. SCHROETER, M. RUDIN
Inst. for Biomed. Engin., ETH, Zuerich, Switzerland

Abstract: Functional magnetic resonance imaging (fMRI) is a technique widely used in research and in clinics, however the biological mechanism underlying the generation of BOLD fMRI signals remains incompletely understood. Astroglial cells are the central elements of the neurovascular unit that link neural circuitry with local blood flow and metabolic support. It has been proposed that release of vasoactive substances by astrocytes couples neuronal activity to changes in cerebrovascular blood flow. However, the contribution of astrocytes to the fMRI BOLD response remains controversial. In order to investigate this question further, we measured simultaneously the calcium activity of astrocytes and neurons in the somatosensory cortex, while

performing fMRI, with the use of genetically encoded calcium indicators (GECIs). This revealed differential time course responses of the two cell types during sensory stimulation, that was diversely correlated with the BOLD signal. In addition, specific activation of astrocytes using DREADDs further contributed to changes in the BOLD signal. Our results point to a novel role of astroglia in the generation of BOLD signal and may further hep the interpretation of fMRI data.

Disclosures: Z. Skachokova: None. **F. Schlegel:** None. **A. Schroeter:** None. **M. Rudin:** None.

Poster

366. Fate Specification and Development of Glia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 366.11/B1

Topic: B.12. Glial Mechanisms

Title: Regulated control of glial cell outgrowth in primary rodent neuronal cultures by CultureOne™ supplement

Authors: M. DERR, D. BEACHAM, N. KAUR, *Y. YAN, D. KUNINGER
Div. of Cell Biol., Thermo Fisher Scientific, Frederick, MD

Abstract: Primary neurons derived from rodent fetal brain are used extensively throughout neuroscience research providing scientists with dynamic and convenient model systems to study basic neuronal function and morphology, disease modeling, drug development, and neurotoxicity. Cryopreserved primary neurons significantly reduce the resources, variability, and time, necessary to isolate these cells from embryos and are routinely used for electrophysiological recording, excitability experiments, and high-throughput and High-Content Screening (HCS) studies. A common complication observed when culturing primary neurons is the presence of contaminating glial cells. The levels of glial cell contamination can vary widely with different isolation methods, age of embryos used, the composition of different media systems, and species of origin. Glial cell contamination and overgrowth can affect assay sensitivity, resolution, and reproducibility. Current methods for reducing glial cell populations in primary neuronal cultures involve treatment with anti-mitotic molecules, such as Cytosine Arabinoside (Ara-C), which have been shown to be toxic to neurons in culture. Gibco CultureOne™ supplement was developed for the differentiation of human pluripotent stem cell-derived neural stem cells (NSCs) into neurons by suppressing the proliferation of NSCs and accelerating the maturation of differentiating neurons. Here we show that addition of CultureOne™ supplement to neuronal culture medium can suppress the outgrowth of contaminating glial cells (astrocytes and oligodendrocytes) in primary rat (E18) cortical neurons, and mouse (E17) cortical and hippocampal neurons. We also demonstrate CultureOne supplement does not impact neuron numbers, morphology, or function. To quantify glial cells

and neurons we employed Immunocytochemistry (ICC) and quantitative image analysis with a High-Content Analysis platform. Depolarization induced calcium influx (Fluo4 Direct assay) and Multi-Electrode Array (MEA) analysis were used to assess neuronal function. Treatment of primary neuron cultures with CultureOne™ supplement at time of cell plating resulted in nearly complete elimination of contaminating glial cells assessed following 14 and 21 days in culture with no significant impact on neuron numbers or morphology. In contrast, delaying the addition of CultureOne™ 3, 7, and 10 days following plating, resulted in increasing levels of glial cells. The results suggest that glial cell levels can be controlled or optimized by adjusting CultureOne™ treatment schedule.

Disclosures: M. Derr: None. **D. Beacham:** None. **N. Kaur:** None. **Y. Yan:** None. **D. Kuninger:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.01/B2

Topic: A.05. Axon and Dendrite Development

Support: KAKENHI 23123519

KAKENHI 24659036

KAKENHI 26460073

KAKENHI 16H01366

KAKENHI 17H03895

AMED ACT-M 17im0210602h0002

AMED ACT-M 16im0210602h0001

Title: Regulation of Reelin function by specific proteolysis

Authors: *M. HATTORI, H. OGINO, E. OKUGAWA, Y. YAMAKAGE, T. KOHNO
Grad. Sch. Pharmaceuti. Sci., Nagoya City Univ., Nagoya, Japan

Abstract: The secreted glycoprotein Reelin regulates embryonic brain development and adult brain functions. It has been suggested that reduced Reelin activity contributes to the pathogenesis of several neuropsychiatric and neurodegenerative disorders, such as schizophrenia and Alzheimer's disease; however, non-invasive methods that can upregulate Reelin activity *in vivo* have yet to be developed. The proteolytic cleavage of Reelin within Reelin repeat 3 (N-t site)

abolishes Reelin activity. We partially purified the enzyme that mediates the N-t cleavage of Reelin from the culture supernatant of cerebral cortical neurons. This enzyme was identified as A Disintegrin And Metalloproteinase with Thrombospondin motifs-3 (ADAMTS-3). Recombinant ADAMTS-3 cleaved Reelin at the N-t site. ADAMTS-3 was expressed in excitatory neurons in the cerebral cortex and hippocampus. N-t cleavage of Reelin was markedly decreased in the embryonic cerebral cortex of ADAMTS-3 knock-out (KO) mice. Importantly, the amount of Dab1 and the phosphorylation level of Tau, which inversely correlate with Reelin activity, were significantly decreased in the cerebral cortex of ADAMTS-3 KO mice. Conditional KO mice, in which ADAMTS-3 was deficient only in the excitatory neurons of the forebrain, showed increased dendritic branching and elongation in the postnatal cerebral cortex. Our study shows that ADAMTS-3 is the major enzyme that cleaves and inactivates Reelin in the brain. Therefore, inhibition of ADAMTS-3 may be an effective treatment for neuropsychiatric and neurodegenerative disorders.
ADAMTS-3 is known to cleave many proteins other than Reelin. Therefore, the phenotypes observed in the brain of ADAMTS-3 KO mice may not derive from the decrease of Reelin N-t cleavage. We thus generated knock-in (KI) mice in which the N-t cleavage site of Reelin had been mutated. The N-t cleavage was dramatically diminished in the brain of the KI mice. More results on these mice will be presented in the meeting.
{References}
Koie et al., Cleavage within Reelin repeat 3 regulates the duration and range of the signaling activity of Reelin protein. **J. Biol. Chem.** 289, 12922 (2014)
Ogino et al., Secreted metalloproteinase ADAMTS-3 inactivates Reelin. **J. Neurosci.** 37, 3181 (2017)

Disclosures: **M. Hattori:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Mitsubishi Tanabe Pharma Corporation. **H. Ogino:** None. **E. Okugawa:** None. **Y. Yamakage:** None. **T. Kohno:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.02/B3

Topic: A.05. Axon and Dendrite Development

Title: Dendritic expansion and spine formation regulated by signaling pathways of unfolded protein response

Authors: *A. SAITO[1], K. IMAIZUMI[2]
[1]Stress Protein Processing, Inst. of Biomed. & Hlth. Sci., [2]Biochemistry, Inst. of Biomed. & Hlth. Sci., Hiroshima Univ., Hiroshima city, Japan

Abstract: The endoplasmic reticulum (ER) is central organelle responsible for calcium ion storage, lipid metabolism, protein synthesis and post-translational modification of secretory and membrane proteins. Various cellular abnormalities such as disturbance of calcium ion homeostasis in ER lumen lead to the disruption of ER function and the accumulation of unfolded proteins (ER stress). The ER stress transducers, IRE1, PERK and ATF6 are activated in response to ER stress, followed by transducing signals from ER to cytoplasm or nucleus to avoid cellular damages (Unfolded Protein Response, UPR). Recent studies have uncovered novel UPR functions not only in dealing with unfolded proteins but also in regulating cellular homeostasis. The well-developed ER network has a highly dynamics that is constantly remodeled and complexly extended from cell soma to distal dendritic segment of neurons, indicating that ER functions could orchestrate local events contributing to dendritic capabilities. In the present study, we focused on the machinery for dendritic functions manipulated by ER-derived signaling including UPR. The phosphorylation levels of IRE1 and PERK were transiently up-regulated by the pre-treatment and washout of primary cultured mouse hippocampal neurons with tetrodotoxin to induce spontaneous excitatory synaptic activity. The activation of these ER stress transducers was observed at post-synaptic sites. We found that the phosphorylation levels were reduced by inhibiting calcium ion outflow from ER, suggesting that the acceleration of calcium ion outflow and its depletion in ER lumen by the excitatory synaptic activation triggers the induction of UPR at post-synaptic sites. The blocking of UPR signaling by the knockdown of UPR-related genes using lentiviruses expressing shRNA inhibited the extension and branching of dendrites. Furthermore, the impaired dendritic spine formation was shown in these knockdown-neurons. Immunofluorescence analysis in the knockdown-neurons revealed that the ER failed to elongate and invaginate into the spine segments. Thus, UPR signaling may regulate the development of intricately branched dendrites and dendritic spine formation through the spatio-temporal fine-tuning of the dendritic ER-dynamics.

Disclosures: A. Saito: None. **K. Imaizumi:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.03/B4

Topic: A.05. Axon and Dendrite Development

Support: DFG Priority Programme SPP 1464

Title: Function of the actin nucleators Spire and Capu in dendrite organisation in *Drosophila*

Authors: *T. STÜRNER[1], H. CUNTZ[2], E. KERKHOFF[3], G. TAVOSANIS[1]
[1]AG Tavosanis, DZNE E.V. Bonn, Bonn, Germany; [2]Ernst Strüngmann Inst. (ESI), Frankfurt, Germany; [3]Univ. Hosp. Regensburg, Regensburg, Germany

Abstract: The correct morphology of dendrites is essential for the function of the nervous system. The underlying cytoskeleton defines the shape and dynamics of dendritic branches under the control of complex protein networks. The aim of this study is to elucidate how regulators of the actin cytoskeleton define the diverse characteristic shapes of dendritic arbors.
Investigating the differentiation of *Drosophila* larva dendritic arborisation (da) neurons, we previously demonstrated that WAVE through recruitment of the Arp 2/3 complex promotes the formation of a branched actin patch at the base of a newly forming branchlet. To elucidate the mechanisms of actin organization following this initial step, we asked whether additional actin nucleating factor are required for branching in these neurons. Here, we focus on the actin nucleators Spire and Capu. While the role of Spire and of the formin Capu in forming a transient actin meshwork during oogenesis has been the object of intensive studies, little is known about the function of these proteins in the nervous system. Spire is negatively regulated by the transcription factor Lola, controlling outgrowth and guidance of axons in the *Drosophila* larval nervous system. In addition, a recent report suggested a role for Spire downstream of Lola also in the establishment of appropriate dendrite morphology of larval da neurons.
We found that Spire and Capu play a role in the formation of the terminal branchlets of Class III da neurons. Class III da neurons have highly actin enriched terminal branchlets and the distinguished dynamics of these branches are essential for the function of the neuron. $Spire^{1/2F}$ or $capu^{1/EE}$ mutant larvae show a reduced number specifically of these characteristic branchlets. Therefore, we analysed this phenotype further by performing *in vivo* time-lapse recordings and are developing quantification methods in the “Trees Toolbox” in Matlab. This data revealed a reduced branchlet extension and retraction in s$pire^{1/2F}$ mutant larvae. Further ongoing studies will elucidate the subcellular localization of these molecules during branch formation and extensions.
Taken together our results suggest a function of Spire and Capu specifically in the formation and dynamics of terminal branchlets of Class III da neurons.

Disclosures: T. Stürner: None. **H. Cuntz:** None. **E. Kerkhoff:** None. **G. Tavosanis:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.04/B5

Topic: A.05. Axon and Dendrite Development

Support: cureCADASIL Research Grant

Davidson FS and R Grant

Title: A novel role for Notch in mechanosensory neuron connectivity in *C. elegans*

Authors: *G. W. BROWN, V, R. EL BEJJANI
Davidson Col., Davidson, NC

Abstract: Notch receptors are conserved transmembrane proteins that regulate key developmental processes and promote stem cell proliferation and renewal. Notch signaling remains active in the nervous system from birth to adulthood. Notch pathway genes are highly conserved across many species, including humans, and extensive work to decipher Notch signaling was initially done in *Caenorhabditis elegans*. While these animals' mechanosensory neurons have been well characterized behaviorally, little is known about the late stage development of the connections between them. We show that animals lacking the Notch metalloprotease *sup-17/*ADAM10 have significantly higher rates of ALM-AVM nerve ring breakage than wild type animals. Significantly higher rates of breakage were found also found in Notch receptor and gamma-secretase complex mutant animals, confirming that the Notch pathway is involved in ALM-AVM connection at the nerve ring. Notch mutants did not exhibit different break rates across life stages. This suggests that the defect is likely to be developmental rather than degenerative or the result of a developmental delay. To ask if Notch functions cell-autonomously, we are currently performing tissue specific rescue experiments for both the mechanosensory neurons and the surrounding glia. We are also assessing the functional role of the nerve ring connection in mechanosensory neurons with optogenetic experiments comparing Notch and wild type worms.

Disclosures: G.W. Brown: None. **R. El Bejjani:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.05/B6

Topic: A.05. Axon and Dendrite Development

Support: Colgate University Research Council

Title: The conserved microtubule-associated protein mini spindles regulates dendrite branching and self-avoidance in *Drosophila* c4da neurons

Authors: *C. KITTOCK, N. ANVERY, M. MISRA
Colgate Univ., Hamilton, NY

Abstract: Dendrite dysgenesis is a common feature of disparate neuropsychiatric and neurodevelopmental disorders, underscoring the need for a more comprehensive understanding of the molecular mechanisms underlying neuronal morphogenesis. The dendrites of Drosophila larval class IV dendritic arborization (c4da) neurons, a subset of cutaneus nociceptive sensory neurons, provide an excellent model system to study the regulation of dendrite morphogenesis because their complex branching patterns show clear parallels to those seen in vertebrates, and their two-dimensional organization on the surface of the larva allows for easy visualization. Recently, a screen for dendrite-localized mRNAs identified the gene *mini spindles* (*msps*) as a positive regulator of dendrite growth. *msps* encodes a microtubule associated protein homologous to XMAP215-family proteins in vertebrates. Previous studies have demonstrated that *msps* regulates microtubule nucleation and promotes dynamic instability. We therefore hypothesized that it might play a role in regulating cytoskeletal events important for the growth and/or retraction of dendrite branches in da neurons during development. RNAi-mediated knockdown of *msps* in c4da neurons resulted in a significant loss of branching and total dendrite length, supporting this initial hypothesis. In addition, reduced *msps* expression resulted in decreased receptive field coverage. Strikingly, a loss of *msps* also impaired intraneuronal dendrite self-avoidance, and $msps^{RNAi}$ neurons exhibited more than twice as many self-crossings as control yw neurons. Self-crossing events have previously been linked to the enclosure of dendrites by overlying epidermal cells; immunostaining for Coracle, a marker of enclosure, confirmed that self-crossing dendrites in $msps^{RNAi}$ neurons also exhibit this behavior. The localization of *msps* mRNA to dendrites and the disorganization of dendrites in the absence of Msps protein preliminarily suggest a model in which local control of *msps* expression could regulate cytoskeletal dynamics in response to signals exchanged between neurons and epidermal cells.

Disclosures: C. Kittock: None. **N. Anvery:** None. **M. Misra:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.06/B7

Topic: A.05. Axon and Dendrite Development

Support: NIH GM103554

Sanford Center for Aging

NIH GM103440

Title: Pink1 regulates brain derived neurotrophic factor signaling to stimulate dendrite outgrowth and mitochondrial function

Authors: *R. K. DAGDA, T. DAS BANERJEE, M. DAGDA, M. SWAIN, R. Y. DAGDA
Pharmacol., Univ. of Nevada Sch. of Med., Reno, NV

Abstract: PTEN-induced kinase 1 (PINK1), which is linked to Parkinson's disease, is a neuroprotective kinase that regulates dendrite remodeling, mitochondrial trafficking and function (Dagda *et al*., 2014, Journal of Neurochemistry, 128(6):864-77). The molecular mechanisms by which PINK1 stimulates dendrite outgrowth remain to be elucidated. Like PINK1, brain-derived neurotrophic factor (BDNF) is a potent stimulator of dendrite outgrowth and maturation. Hence, we surmised that PINK1 regulates dendrite outgrowth via BDNF. Immunohistochemical analyses of brain slices derived from 10 month old PINK1 knockout mice show that PINK1-deficient cortical and hippocampal neurons have reduced intracellular levels of BDNF and a concomitant reduction in dendrite length. *In vitro*, time-dependent analyses of dendrite length and complexity revealed that PINK1-deficient primary neurons exhibit significantly decreased outgrowth rates and retraction of dendrites upon reaching maturation in culture. Treating PINK1-deficient neurons with recombinant human BDNF was able to restore dendrite length in PINK1-deficient cortical neurons to similar levels as wild-type neurons. Conversely, treating PINK1-overexpressing SH-SY5Y cells with inhibitors of the BDNF receptor (TrkB) blocked the ability of PINK1 to enhance neurite outgrowth. These results suggest that PINK1 stimulates dendrite outgrowth through BDNF-mediated activation of TrkB. Mechanistically, PINK1 activates PKA signaling to enhance BDNF levels as transfecting neuroblastoma SH-SY5Y cells with an inhibitor of Protein Kinase A (PKI) was able to block PINK1's ability to enhance intracellular levels of BDNF. Primary neurons treated with human recombinant BDNF (2-24 hours) showed enhanced oxygen consumption rates (basal, maximal and ATP-linked OCRs), increased rates of glycolysis, increased anterograde mitochondrial trafficking, increased mitochondrial content and fusion suggesting that BDNF phenocopies the ability of PINK1 to enhance the bioenergetics status of the cell. Overall, our data suggest the existence of a new neuroprotective signaling axis in which PINK1 stimulates BDNF by activating PKA signaling to modulate dendrite outgrowth and enhance mitochondrial function in neurons.

Disclosures: R.K. Dagda: None. **T. Das Banerjee:** None. **M. Dagda:** None. **M. Swain:** None. **R.Y. Dagda:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.07/B8

Topic: A.05. Axon and Dendrite Development

Support: KAKENHI 26460073

KAKENHI 17H03895

AMED ACT-M 17im0210602h0002

AMED ACT-M 16im0210602h0001

Title: Contribution of ADMATS family members to Reelin inactivation in the postnatal brain

Authors: *M. KATO[1], T. KOHNO[2], M. HATTORI[2]
[1]Grad. Sch. Pharmaceuti. Sci., Nagoya City Univ., Nagoya-Shi, Japan; [2]Grad. Sch. Pharmaceuti. Sci., Nagoya City Univ., Nagoya, Japan

Abstract: Reelin is a large secreted protein that is essential for normal brain development. In the postnatal brain, Reelin plays important roles in modulating synaptic plasticity and functions. Accordingly, downregulation and hypoactivity of Reelin in the postnatal brain have been suggested to be involved in the pathogenesis of several neuropsychiatric and neurodegenerative disorders, such as Alzheimer's disease and schizophrenia. Therefore, upregulation of Reelin activity can be a therapeutic strategy for these disorders. Reelin binds to its cell-membrane receptors and induces phosphorylation and subsequent degradation of intracellular protein Dab1. A specific cleavage of Reelin, called N-t site cleavage, virtually abolishes Reelin's ability to induce phosphorylation and degradation of Dab1 (Kohno et al., 2009). Previously, we identified ADAMTS-3 (A disintegrin and metalloproteinase with thrombospondin motifs 3) as the major proteinase in charge of the N-t site cleavage in the embryonic and the early postnatal brain (Ogino et al., 2017). However, it was also suggested that other proteinase(s) may contribute to N-t site cleavage in the postnatal brain. The domain structure of ADAMTS-3 is almost identical to that of ADAMTS-2 and -14, and these three molecules compose the subfamily of ADAMTS family members called procollagen N-proteinases, suggesting that ADAMTS-2 and -14 may also cleave Reelin at the N-t site. The aim of this study is to clarify the contribution of ADAMTS-2 and -14 to the N-t site cleavage of Reelin in the postnatal brain. The recombinant ADAMTS-2 protein had the ability to cleave Reelin at the N-t site, whereas ADAMTS-14 did not. The mRNA of ADAMTS-2 was expressed in the postnatal brain, whereas ADAMTS-14 was not. These results suggested that ADAMTS-2 is the good candidate proteinase in charge of N-t site cleavage in the postnatal brain. We then generated ADAMTS-2 knock-out mice by using the CRISPR/Cas9 system. We are now analyzing these mice to understand the contribution of ADAMTS-2 to the N-t site cleavage of Reelin.

Disclosures: **M. Kato:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; KAKENHI 26460073, KAKENHI 17H03895, AMED ACT-M 17im0210602h002, AMED ACT-M 16im0210602h0001. **T. Kohno:** None. **M. Hattori:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Mitsubishi Tanabe Pharma Corp..

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.08/DP01/B9 (Dynamic Poster)

Topic: A.05. Axon and Dendrite Development

Support: JSPS Grant-in-Aid for Scientific Research

Title: *In vivo* dendritic development of cerebellar Purkinje cells

Authors: *Y. TAKEO, E. MIURA, M. YUZAKI
Dept. of Neurophysiol., Keio Univ. Sch. of Med., Tokyo, Japan

Abstract: Neurons exhibit a specialized dendritic morphology, which is specific to the neuronal types and closely related to their function in the neural circuit. However, molecular mechanisms underlying *in vivo* dendritic development are largely unknown. Cerebellar Purkinje cells (PCs) have a unique dendritic architecture characterized by a single primary dendrite with a finely branched, flat fan-like arborization. These features of dendrites are established through dynamic morphological remodeling during early postnatal ages. At birth, post-migratory PCs exhibit fusiform shape with long primitive dendrites. In the next few days, these primitive dendrites are retracted, and the PCs enter the “stellate cell” stage with multiple short dendrites radiated in all directions. Shortly thereafter, by around P8 in mice, the PCs suddenly turn into the “young PC” stage equipped with the typical single stem dendrites and flatten branches. The molecular mechanism of this remodeling process from the “stellate-cell” stage to the “young PC” stage has been one of the great mysteries of dendritic morphogenesis. To address this question, we investigated how single stem dendrites and flatten branches are generated by *in vivo* two-photon imaging of developing PCs in mouse cerebellum. We revealed that dendrites actively change their length and branching patterns during transition from the “stellate-cell” stage to the “young PC” stage. We also found that the neuronal activity of PCs is required for this process by overexpression of the inwardly rectifying potassium channel (Kir2.1) *in vivo*. Our study shed light on how a cell-type specific three-dimensional dendritic morphology is established *in vivo*.

Disclosures: Y. Takeo: None. **E. Miura:** None. **M. Yuzaki:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.09/B10

Topic: A.05. Axon and Dendrite Development

Support: NIH/NINDS R01NS083947

Title: Premature birth reduces CA1 hippocampal dendrite branching, spine density, and neurocognitive function

Authors: D. KLEBE[1], M. TIBREWAL[4], B. CHENG[1], P. DOHARE[1], P. R. MOUTON[5], K. DOBRENIS[2], J. VELISKOVA[4], *P. BALLABH[3]
[2]Neurosci., [3]Pediatrics and Neurosci., [1]Albert Einstein Col. of Med., Bronx, NY; [4]New York Med. Col., Valhalla, NY; [5]Pathology & Cell Biol., USF Hlth. Byrd Alzheimer's Inst., Tampa, FL

Abstract: Preterm-born children and adolescents suffer neurological and behavioral disorders. These neurobehavioral conditions might be attributed to the effect of premature birth, suboptimal nutritional support or rearing practices (breast vs. formula feeding) on brain development. The hippocampus plays distinctive roles in memory, learning, flexible cognition, and social behavior. Therefore, we hypothesized that premature birth and formula feeding of preterm newborns might impact neurobehavioral function, hippocampal dendritic arborization, and density of dendritic spines. To test our hypotheses, we compared preterm (E28.5) and term (E32) rabbit pups at an equivalent post-conceptional age (P28) for neurobehavioral function and dendritic length, branching, and spines in the CA1 area of the hippocampus. Preterm pups were reared in an infant incubator and were gavage fed, whereas term pups were reared by mother rabbit. Neurobehavioral tests included open field test, novel object recognition (NOR; recognition memory), elevated plus maze, three chambered socialization test, and Barnes Maze test, using video tracking system (ANY-maze, Stoelting Co.). Dendritic length, branching, and spines in pyramidal neurons of the CA1 area of the hippocampus were evaluated in Golgi-stained sections and all the quantifications were done by computerized stereology (Stereologer, SRC Biosciences, Tampa, FL). We found that preterm pups spent more time in arena center compared to term pups ($P<0.05$). The speed of locomotion and distance travelled in the arena were comparable between two groups. Preterm pups spent less time with the novel object compared to term pups ($P<0.05$). Three chambered socialization test revealed that preterm pups showed less interest in the stranger compared to term pups ($P<0.05$). Dendritic branching and dendritic spine density were significantly reduced in preterm rabbits compared to term controls ($P=0.025$ and 0.037). Dendritic length showed a non-significant decrease in preterm rabbits compared to controls. These data suggest preterm rabbits lack preference for novelty and display poor learning and memory, which can be ascribed to reduced dendritic arborization and fewer dendritic spines in

the CA1 area of the hippocampus. Premature birth and postnatal rearing practices in humans might result in enduring microstructural changes in brain growth and development.

Disclosures: D. Klebe: None. **M. Tibrewal:** None. **B. Cheng:** None. **P. Dohare:** None. **P.R. Mouton:** None. **K. Dobrenis:** None. **J. Veliskova:** None. **P. Ballabh:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.10/B11

Topic: A.05. Axon and Dendrite Development

Support: NIH R21 NS090030

NIH R01 NS055272

Title: The γ-Protocadherins regulate dendrite arborization in the cerebral cortex through both common and isoform-specific intracellular signaling mechanisms

Authors: K. MAH[1], A. M. GARRETT[2], R. W. BURGESS[3], *J. A. WEINER[4]
[1]Biol., Univ. of Iowa, Iowa City, IA; [2]The Jackson Lab., Bar Harbor, ME; [3]Jackson Lab., Bar Harbor, ME; [4]Dept. of Biol., The Univ. of Iowa, Iowa City, IA

Abstract: A key component of neural circuit formation is the elaboration of complex dendritic arbors, the pattern of which constrains inputs to the neuron and thus the information it processes. As such, many neurodevelopmental disorders such as autism and Down, Rett, and Fragile X Syndromes are associated with reduced forebrain dendrite arborization. The *Pcdhg* gene cluster encodes a family of 22 homophilic cell adhesion molecules, the γ-Protocadherins (γ-Pcdhs), that we have shown are critical for the elaboration of complex dendrite arbors in the cerebral cortex. Each γ-Pcdh isoform is unique in its extracellular and proximal cytoplasmic domain, but all 22 γ-Pcdhs share the same C-terminal constant domain. We recently found that PKC phosphorylation at a serine residue in the constant domain prevents the γ-Pcdhs from promoting dendrite complexity through inhibition of FAK in cultured neurons (Keeler et al., *JBC* 2015). In a separate study, we found that the unique cytoplasmic domain of one isoform, γ-Pcdh-C3, can inhibit Wnt signaling in HEK293 cells through its interaction with and stabilization of Axin1, a protein also necessary for dendrite arborization (Mah et al., *Sci. Rep.* 2016). Here, we have utilized CRISPR/Cas9 genome editing to generate 3 new mouse lines: 1) $Pcdhg^{S/A}$, in which the PKC target serine residue in the constant domain is mutated to an alanine, preventing PKC phosphorylation of all γ-Pcdhs; 2) $Pcdhg^{CTD}$, in which an early stop codon results in all γ-Pcdhs lacking the C-terminal 15 amino acids of the constant domain (including the PKC target serine); and 3) $Pcdhg^{C3KO}$, in which a 13 bp deletion near the ATG of the *PcdhgC3* variable exon

generates a null for this isoform. All mice are viable and fertile, and the expected protein modifications are observed: γ-Pcdhs are entirely unphosphorylated at the C-terminal serine in both *Pcdhg*$^{S/A}$ and *Pcdhg*^{CTD}, and the C3 isoform (and only this isoform) is absent in *Pcdhg*^{C3KO}. We utilized *Thy1-YFPH* mice to label cortical layer V pyramidal neurons and analyzed dendrite arborization at 3 and 6 weeks of age. Arborization was significantly altered in all 3 lines: an increase in arbor complexity in both *Pcdhg*$^{S/A}$ and *Pcdhg*^{CTD} animals, and a decrease in complexity in *Pcdhg*^{C3KO} mice. As predicted, we also observe increased canonical Wnt signaling in *Pcdhg*^{C3KO} brain, assayed using a Wnt reporter allele; current experiments are aimed at confirming Axin1 as an effector of γ-Pcdh-C3's promotion of dendrite complexity *in vivo.* These observations confirm that γ-Pcdhs can regulate dendrite arborization through multiple intracellular pathways, and provide new *in vivo* tools for the genetic dissection of Pcdh mechanisms of action.

Disclosures: K. Mah: None. **A.M. Garrett:** None. **R.W. Burgess:** None. **J.A. Weiner:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.11/B12

Topic: A.05. Axon and Dendrite Development

Support: NARSAD 2012 Marion G. Nicholson Distinguished Investigator Award

NSF grant IOS-1353724

NIH Biotechnology Training Grant 5 T32 GM 8339-27

Title: Effects of antipsychotics and NMDA receptor agonists on expression and function of NOS1AP, a protein encoded by a schizophrenia susceptibility gene

Authors: *K. SVANE, E. ASIS, B. L. FIRESTEIN
Rutgers Univ., Piscataway, NJ

Abstract: Schizophrenia is a debilitating mental illness that affects 1% of the U.S. population and includes positive, negative, and cognitive symptoms. Current antipsychotics target dopamine signaling and are unable to treat all symptom domains of the disorder. Therefore effective treatments for schizophrenia represent a significant unmet medical need. *N*-methyl D-aspartate (NMDA) receptor signaling has also been implicated in the pathophysiology of schizophrenia and NMDA receptor agonists represent promising potential treatment options. Nitric Oxide Synthase 1 Adaptor Protein (NOS1AP) is a protein encoded by a schizophrenia susceptibility gene that negatively regulates NMDA receptor signaling. Moreover, NOS1AP protein is overexpressed in the dorsolateral prefrontal cortex (DLPFC) of postmortem tissue of patients

with schizophrenia and has been found to reduce dendrite branching, which is also observed in individuals with schizophrenia. Here, we found that the NMDA receptor agonist D-serine is able to significantly reduce NOS1AP protein expression in embryonic rat cortical neurons and is able to rescue NOS1AP-mediated reductions in dendrite branching. On the other hand, traditional antipsychotics such as clozapine, haloperidol, and fluphenazine have no effect on NOS1AP expression or dendrite branching. Ultimately, our research will shed light on the therapeutic potential of D-serine as a treatment for schizophrenia and the mechanism by which D-serine takes effect.

Disclosures: K. Svane: None. **E. Asis:** None. **B.L. Firestein:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.12/B13

Topic: A.05. Axon and Dendrite Development

Support: National Research Centre grant no. 2011/03/B/NZ3/01970

Title: The role of adaptor complex AP2 in formation of dendritic arbors of hippocampal neurons

Authors: A. KOSCIELNY, A. MALIK, E. LISZEWSKA, J. ZMORZYNSKA, A. TEMPES, B. TARKOWSKI, *J. JAWORSKI
Intl. Inst. of Mol. and Cell Biol., Warsaw, Poland

Abstract: Introduction: The proper dendritic branching is a highly regulated process. Among its regulators are membrane proteins internalized via clathrin-mediated endocytosis. AP2 adaptor complex is a key player in this process, but its role in mammalian dendritogenesis has not yet been tested. **Aim:** The aim of this study was to find how AP2 complex contributes to shaping dendritic tree of developing hippocampal neurons. **Methods:** To study role of AP2 complex in dendritic arborization we used primary hippocampal neurons expressing AP2b1 (β-adaptin) shRNA alone or in combination of functional rescue constructs (i.a. GluA2, $S6K1^{ca}$). The effect was also tested *in vivo* by lentiviral injections to newborn rats. Upon β-adaptin knockdown, we tested GluA2 trafficking via internalization assay and GluA2 level by Western blot and immunochemistry. GluA2 degradation and mTOR dependent biosynthesis were investigated by e.g. cycloheximide or rapamycin treatment. **Results:** We showed that knockdown of β-adaptin led to reduction in dendritic arbors of developing hippocampal neurons *in vitro* and *in vivo*. The knockdown of AP2 also led to decreased level of GluA2, what is a result of impaired mTOR dependent GluA2 biosynthesis. However, the overexpression of functional GluA2 or restoration of mTOR activity rescued this effect. **Conclusions:** AP2 adaptor complex regulates the dendritogenesis of mammalian neurons via mTOR dependent GluA2 biosynthesis.

Disclosures: **A. Koscielny:** None. **A. Malik:** None. **E. Liszewska:** None. **J. Zmorzynska:** None. **A. Tempes:** None. **B. Tarkowski:** None. **J. Jaworski:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.13/B14

Topic: A.05. Axon and Dendrite Development

Support: NIH Grant R01EY024694

Pew Charitable Trusts

McKnight Endowment Fund for Neuroscience

Title: Identifying new molecular cues that guide development of retinal direction-selective circuitry

Authors: *C. L. PRIGGE, J. N. KAY
Neurobio., Duke Univ., Durham, NC

Abstract: During development, some neurons that share a role in neural processing arborize in the same synaptic layers, avoiding inappropriate layers, implying neurons can distinguish between each other—a mechanism likely involving molecular recognition cues. We seek to discover molecules that guide the selective lamination and wiring between neurons of the mouse retinal direction-selective (DS) circuit. This well-characterized circuit comprises three cell types—DS bipolar cells, starburst amacrine cells, and DS ganglion cells—that project to two specific layers in the inner plexiform layer (IPL). Because these cells can be genetically manipulated and marked, we can study how their selective connections arise. To identify candidate molecules that mediate laminar recognition in this circuit, we queried our microarray dataset which contains gene expression profiles of 19 retinal cell types at postnatal day 6, an important time for retinal synaptogenesis. We selected genes predicted to encode cell-surface or secreted proteins, which could act as guidance cues. Secondary screens for molecular interactions among candidates were also performed, and the data considered when selecting candidates. Here we focus on two candidates, *Tgfb3* and *Unc5c*. *Tgfb3*, which encodes a transforming growth factor beta signaling protein, was enriched in starbursts. TGF-beta superfamily members have broad roles in signaling and development, including regulation of extracellular matrix and synapse formation in fly neuromuscular junction. *Tgfb3* is therefore well positioned to act as a circuit-specific ligand promoting laminar targeting or synapse formation. *Unc5c*, which encodes a repulsive guidance receptor, is excluded from the DS circuit, but one of its ligands, FLRT2, is localized to the circuit, suggesting it might function in laminar restriction

of DS circuit arbors. We characterized the expression patterns of these molecules to confirm selective expression. We found that *Tgfb3* mRNA and protein were exclusive to starbursts. *Unc5c* mRNA colocalized with the majority of GAD65+ GABAergic cells, a population which avoids the DS layers. To ask if these molecules are required for DS circuit development we used Hb9-GFP mice (which mark DS ganglion cells) and ChAT-Cre mice (a starburst-specific Cre) to examine DS neuron IPL laminar targeting and single cell morphology in mutant mice. Sporadic laminar targeting errors were observed in both *Tgfb3* and *Unc5c* mutants. Altered starburst dendritic branching in *Tgfb3* mutants suggests synaptic connectivity may be affected. We are now investigating the role that these molecules may play in DS circuit laminar targeting and synapse formation.

Disclosures: C.L. Prigge: None. **J.N. Kay:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.14/B15

Topic: A.05. Axon and Dendrite Development

Support: NIH Grant R01EY024694

McKnight Endowment Fund for Neuroscience

Pew CharitableTrusts

Title: Characterizing cell surface receptor multimeric complexes for homotypic recognition in developing retinal circuits

Authors: *J. WANG, J. N. KAY
Duke Univ., Durham, NC

Abstract: In the central nervous system, billions of neurons interconnect with precision to form morphologically complex and functionally diversified neural circuits. The stereotypical fashion by which neurons assemble suggests that cell-surface molecular cues can act as identity tags during development. In many cases, cell surface receptors bind ligands expressed on other cells in order to distinguish homotypic neighbors. Two closely-related transmembrane receptors containing multiple epidermal growth factor (EGF)-like domains, *Megf10* and *Megf11*, were previously identified to mediate homotypic recognition of certain retinal cell types. Genetic evidence suggests that MEGF10 acts as both ligand and receptor to initiate cell-cell repulsion. MEGF11 can also act as a ligand, but even though retinal neurons express both molecules, genetic evidence suggests it does not act through MEGF10. To complicate things further, MEGF10 and 11 ligands and their receptors are co-expressed in the same neurons during

development. These observations raise many questions about the biochemical basis of the receptor and ligand signaling complex. If these complexes are multimeric, selective *cis* interactions between MEGF10 and 11 might generate complexes with distinct properties, thereby increasing the complexity of homophilic interactions. To address this question, we first tested whether MEGF10 can interact homophilically in *cis*. We expressed MEGF10 with two epitope tags in the same cells. Through co-immunoprecipitation (IP), we discovered that MEGF10 indeed interacts. Since MEGF10 does interact in *cis* configuration, which domain(s) of MEGF10 is responsible for such binding? First we deleted the entire intercellular domain and replaced it with epitope tags. Co-IP demonstrated that MEGF10 *cis* interactions likely occur through its ectodomain. To discover the crucial regions responsible for this interaction, extracellular domain truncations were constructed. Similar experiments are underway for MEGF11. Once we have mapped the interaction region(s) responsible for *cis* interactions, these can be used for *in vivo* functional study to evaluate the biological importance of such interaction. Our results set the stage to uncover the molecular mechanism by which MEGF10 and 11 mediate cell-cell recognition.

Disclosures: J. Wang: None. **J.N. Kay:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.15/B16

Topic: A.05. Axon and Dendrite Development

Support: NIH

Title: Neuron dendrograms are produced by an asymmetrical process

Authors: *R. FARHOUDI[1], D. ROLNICK[2], P. RAMKUMAR[3], K. P. KORDING[4]
[1]Mathematics, Sharif Univ., Tehran, Iran, Islamic Republic of; [2]Mathematics, Massachusett Inst. of Technol., Boston, MA; [3]Physical Med. & Rehabil. and Neurobio., [4]Physiol& PMnR, Northwestern Univ., Chicago, IL

Abstract: Neuroscience has compiled huge databases of neuron morphologies, and describing their structure is thus an exciting scientific topic. A morphology consists of many aspects, including segmental shapes, angular relations across segments, and the topological dendrogram. The established growth model for dendrograms is the Galton-Watson process, which splits a branch with a certain probability and otherwise produces a leaf node. We find that it cannot be improved by adding depth dependency of the probability. We find, however, that it can be improved with a simple asymmetrical growth model. In this model, each split produces two different edges, where each of them has a different probability of branching vs. dying. This

model considerably outperforms the other tested models over our dataset of around 60000 trees. Our result suggests that the generative process for neuron dendrograms exhibits asymmetrical processes.

Disclosures: **R. Farhoudi:** None. **D. Rolnick:** None. **P. Ramkumar:** None. **K.P. Kording:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.16/B17

Topic: A.05. Axon and Dendrite Development

Support: NIH R01 HD029421

NIH F32 HD085576

Title: Choline supplementation ameliorates dendrite complexity impairments in developing iron-deficient hippocampal neurons

Authors: *T. W. BASTIAN[1,2], W. C. VON HOHENBERG[1], L. M. LANIER[2], M. K. GEORGIEFF[1]
[1]Pediatrics, [2]Neurosci., Univ. of Minnesota, Minneapolis, MN

Abstract: Iron deficiency (ID), with or without anemia, affects an estimated 2 billion people worldwide. ID is particularly deleterious during fetal/neonatal brain development, leading to neurological impairments, including deficits in hippocampal-mediated learning and memory. These deficits often persist into adolescence and adulthood despite early-life iron repletion. Unfortunately, in some iron-deficient populations nutritional iron supplementation is either not realistic or can even be dangerous (e.g., in malaria endemic countries). Because iron therapy is not always possible or effective, it is necessary to develop alternative treatment strategies targeting the underlying neurodevelopmental deficits of early-life ID. Supplementation with choline in conjunction with iron repletion therapy improves adult rat learning and memory following recovery from early-life ID.
The cellular basis of choline's restorative effect on ID-induced learning and memory impairments was determined using embryonic hippocampal neuronal cultures made iron-deficient by adding 10 μM deferoxamine (DFO, an iron chelator) beginning at 3 days in vitro (DIV). At 11 DIV, iron-deficient cultures were treated with iron, choline (30 μM), iron/choline, or left untreated. At 18 DIV, the dendritic arbors for individual neurons were manually traced and Sholl analysis was performed to assess dendritic arborization.
Consistent with our previous findings, DFO treatment significantly reduced complexity

throughout the dendritic arbor by reducing the length, but not the number, of primary dendrites and branches. Choline treatment, alone or in combination with iron, recovered dendritic arbor complexity, including dendrite and branch lengths to levels indistinguishable from control or iron-repleted neurons. Our findings show, for the first time, that choline acts directly on neurons and does not require iron repletion to mediate its restorative effect following early-life ID. While the molecular mechanism(s) of choline action during early-life ID remain unclear, choline may have clinical potential for iron-deficient children when iron supplementation is not advised.

Disclosures: T.W. Bastian: None. **W.C. von Hohenberg:** None. **L.M. Lanier:** None. **M.K. Georgieff:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.17/B18

Topic: A.05. Axon and Dendrite Development

Title: Investigating the role of Csmd2 in Reelin-mediated dendrite formation and synaptogenesis

Authors: *M. A. GUTIERREZ[1], S. J. FRANCO[2]
[1]Univ. of Colorado Denver, Anschutz Med., Aurora, CO; [2]Pediatrics, Univ. of Colorado Sch. of Med., Aurora, CO

Abstract: Reelin is a secreted glycoprotein that regulates development of the cerebral cortex. By initiating a signaling cascade through the downstream adaptor protein, Dab1, Reelin has been extensively studied as a migration guidance cue for immature neurons in the developing brain. Emerging evidence indicates that Reelin signaling continues to play a key role in mature neurons, as it is important for dendritic branching and synapse formation, thereby affecting learning and memory. However, the mechanisms downstream of Reelin and Dab1 that regulate these functions are not currently known. Here, we have identified a novel interaction between Dab1 and Csmd2, a single-pass transmembrane protein of unknown function. We demonstrate that Dab1 binds to the FENPxY motif in the cytoplasmic tail of Csmd2. Additionally, we show that Csmd2 is required for Reelin-mediated enhancement of dendritic branching in cultured hippocampal neurons. Interestingly, we find that Csmd2 also interacts with several synaptic scaffolding proteins and localizes to synapses in vitro and in vivo. Furthermore, knockdown of Csmd2 mRNA or expression of a truncated form of Csmd2 in mouse cortical and hippocampal neurons causes a significant reduction in dendritic spine number and density. Together, these data indicate a role for Csmd2 in dendritogenesis and synapse formation, and suggest that Csmd2 may be a critical factor downstream of Reelin/Dab1 during neuronal maturation and differentiation.

Disclosures: **M.A. Gutierrez:** None. **S.J. Franco:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.18/B19

Topic: A.05. Axon and Dendrite Development

Support: National Research Foundation of Korea (NRF)

Title: A penetrable nuclear/nucleolar protein, mLLP, is regulator of neural development and its synaptic transmission

Authors: ***S.-W. LEE**[1], N.-K. YU[2], H. KIM[3], J. SHIM[1], S. KIM[1], D. KIM[4], C. KWAK[1], S.-E. SIM[1], J.-H. CHOI[1], S. AHN[1], J. YOO[1], S.-L. CHOI[1], D.-J. JANG[5], C.-S. LIM[1], Y.-S. LEE[6], C. KANG[7], S. CHOI[4], B.-K. KAANG[1]
[1]Seoul Natl. Univ., Seoul, Korea, Republic of; [2]UCSD, LA Jolla, CA; [3]Sungkyunkwan Univ., Suwon, Korea, Republic of; [4]Hallym Univ., Chunchon, Korea, Republic of; [5]Kyungpook Natl. Univ., Sangju-si/gyeongsangbuk-Do, Korea, Republic of; [6]Chung-Ang Univ., Seoul, Korea, Republic of; [7]Kyung Hee Univ., Suwon, Korea, Republic of

Abstract: Mouse LLP homolog (mLLP) is a novel cell-permeable protein and it has roles in regulation of neural development as nuclear/nucleolar protein. Following our experiment, mLLP is strongly expressed in neural development, and modulation of mLLP level during maturation of cultured neurons affected synaptic transmission and neuronal growth. In addition, ectopic overexpression of mLLP protein increased dendritic arborization, demonstrating the non-cell-autonomous effect of mLLP. Futhermore, we found upstream and downstream components of mLLP, and also found that CCCTC-binding factor (CTCF) as well as transcriptional regulation factor interacts with mLLP. This interaction can modulate gene expression involved in neuronal growth. Taken together, these results demonstrate that mLLP has important roles in modulating neural development, and it can affect synaptic transmission and neuronal growth.

Disclosures: **S. Lee:** None. **N. Yu:** None. **H. Kim:** None. **J. Shim:** None. **S. Kim:** None. **D. Kim:** None. **C. Kwak:** None. **S. Sim:** None. **J. Choi:** None. **S. Ahn:** None. **J. Yoo:** None. **S. Choi:** None. **D. Jang:** None. **C. Lim:** None. **Y. Lee:** None. **C. Kang:** None. **S. Choi:** None. **B. Kaang:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.19/B20

Topic: A.05. Axon and Dendrite Development

Support: NIH Grant NS090030

NIH Grant NS054154

Title: Neurodevelopmental consequences of reducing gamma-protocadherin isoform diversity with CRISPR/Cas9 genome editing

Authors: *A. M. GARRETT[1], J. A. WEINER[2], R. W. BURGESS[1]
[1]The Jackson Lab., Bar Harbor, ME; [2]Dept. of Biol., The Univ. of Iowa, Iowa City, IA

Abstract: The mammalian *Pcdhg* gene cluster on human chromosome 5q31 and mouse chromosome 18 consists of 22 variable exons and 3 constant exons. Promoter choice mechanisms involving differential methylation and DNA looping generate transcripts encoding the gamma-protocadherins (γ-Pcdhs), a family of 22 distinct cadherin superfamily cell adhesion molecule isoforms. The majority of each isoform (6 extracellular cadherin repeats, a transmembrane domain, and a variable cytoplasmic domain) is encoded by a single variable exon, while a common cytoplasmic C-terminus is encoded by the constant exons. The γ-Pcdhs are expressed broadly throughout the nervous system, with individual cells expressing ~7 of the 22 isoforms, including ~4 of the stochastically expressed gamma-A and –B subfamily genes plus the 3 ubiquitously expressed gamma-C subfamily genes. The proteins form combinatorial multimers promiscuously in *cis*, but the binding specificity across membranes in *trans* is strictly homophilic at the multimer level. Thus, through isoform combination, the γ-Pcdhs themselves could generate thousands of distinct recognition units; given that γ-Pcdhs can also be found in complexes with the α- and β-Pcdhs, encoded by adjacent *Pcdha* and *Pcdhb* gene clusters, possibly billions. The γ-Pcdhs critically regulate multiple neurodevelopmental processes, including synapse formation, neuronal survival, dendrite self-avoidance, and dendrite arborization, but the role of isoform diversity in these functions is still unclear. Previous experiments testing isoform variety used the relatively blunt tools of complete cluster knockouts and single isoform transgene overexpression. To more precisely ask whether molecular diversity is essential for normal γ-Pcdh functions, we used CRISPR/Cas9 genome editing to generate an array of mutant and internal deletion alleles in the *Pcdhg* locus in mice. Single guide RNAs were designed to target sequences near the ATG of each of the 22 *Pcdhg* variable exons, and were injected together as a mixture into fertilized eggs. We used an Illumina TruSeq Custom Amplicon panel to sequence the targeted genomic regions and the top predicted off-target sites in the resultant mice, and established 30 new lines with between 1 and 16 disrupted variable exons.

Multiple lines with substantially reduced isoform diversity are viable as homozygotes, despite the neonatal lethality of *Pcdhg* cluster null mice. Currently, we are generating homozygous mutants for additional lines to analyze neurodevelopmental phenotypes, and are using whole genome sequencing to validate mutations.

Disclosures: A.M. Garrett: None. **J.A. Weiner:** None. **R.W. Burgess:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.20/B21

Topic: A.05. Axon and Dendrite Development

Support: The Danish Council for Independent Research

Title: Early life vitamin C deficiency does not alter CA1 neuronal morphology or markers of synaptic plasticity in a guinea pig model

Authors: *S. N. HANSEN[1], J. R. NYENGAARD[2], J. LYKKESFELDT[1], P. TVEDEN-NYBORG[1]
[1]Dept. of Vet. and Animal Sci., Univ. of Copenhagen, Frederiksberg C, Denmark; [2]Dept. of Clin. Med., Aarhus Univ., Aarhus, Denmark

Abstract: Background: Vitamin C (VitC) deficiency affects up to 10 % of the population including children. VitC is highly concentrated in the brain, acting as an essential antioxidant and partaking in monoaminergic neurotransmitter synthesis and glutamate reuptake. We have shown that early life VitC deficiency impairs spatial memory and reduces hippocampal (HP) neuron numbers and maturation and HP volume in guinea pigs. Neuronal morphology and synaptic plasticity are crucial in establishing functional circuits in the brain, thus impairments could likely reduce signal transmission and decrease functionality. As a precocial species and one of few mammalian species unable to synthesize VitC, guinea pigs - like humans- depend exclusively on a dietary supply, making them a unique *in vivo* model within this specific field.
Objective: To investigate if early life VitC deficiency impairs the morphology of hippocampal *cornu ammonis* 1 (CA1) pyramidal neurons and reduces the expression of molecular markers of synaptic plasticity in HP, frontal cortex (FC) and striatum (S).
Methods: Seven days old guinea pigs (n=30) were block randomized to three weight stratified groups receiving either: 1500 mg VitC/kg diet (CTRL), 100 mg VitC/kg diet (Def) or 0 (first two weeks) to 50 mg VitC/kg diet (Sev-def) for eleven weeks. The right hemispheres from CTRL and Sev-def animals were Golgi-stained and Z-stacks from 5-6 neurons from CA1 of the HP were reconstructed in 3D and analyzed. Left hemisphere HP, FC and S were analyzed for markers of synaptic plasticity.

Results: The Def and Sev-def groups showed mild growth retardation, but showed no difference in adjusted brain weight. Cortical VitC levels were reduced in Def and Sev-def. Neither apical nor basal CA1 neuronal dendrites or spines were affected by VitC status. Plasticity markers, brain-derived neurotrophic facor, p-/synapsin-1, p-/ Ca^{2+}/calmodulin-dependent protein kinase II, did not significantly differ.
Conclusion: Early life VitC did not compromise neuronal morphology in the CA1 and caused no regional changes in the markers of synaptic plasticity. Whether VitC deficiency alters other markers and regions, including astrocyte VitC recycling, should be explored further.

Disclosures: S.N. Hansen: None. **J.R. Nyengaard:** None. **J. Lykkesfeldt:** None. **P. Tveden-Nyborg:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.21/B22

Topic: A.05. Axon and Dendrite Development

Support: Wagner Fellowship

NIH Grant F31NS098621

Double-Hoo Research Grant

NESC Training Grant

Title: Trafficking of the TrkA signaling endosome

Authors: *K. A. BARFORD[1], K. MCDANIEL[1], C. YAP[3], C. DEPPMANN[2], B. R. WINCKLER[4]
[2]Biol., [1]Univ. of Virginia, Charlottesville, VA; [3]Cell Biol., Univ. of Virginia Sch. of Med., Charlottesville, VA; [4]Dept Neurosci, Univ. of Virginia Dept. of Neurosci., Charlottesville, VA

Abstract: Protein trafficking is involved in all aspects of neuronal function including development, axon/dendrite growth, and synaptic function. Neurons have distinct trafficking challenges because of the extreme length of their axons and dendrites. For this reason, neurons express special endosomal machinery to regulate protein trafficking. Peripheral neurons, including sympathetic neurons, are among those in distinct need of specialized protein transport due to their long axons. One receptor that is critical for the development of the sympathetic nervous system, TrkA, has been shown to undergo complex trafficking events. In sympathetic neurons, TrkA is expressed during development and encounters its high-affinity ligand, Nerve Growth Factor (NGF), once the axon reaches its final target tissue. TrkA binds NGF and the

complex is internalized into a signaling endosome. The NGF-TrkA signaling endosome is critical in the axon for outgrowth and branching, but it is also transported retrogradely to the soma and to dendrites where it is necessary for survival signaling and synapse formation, respectively. We hypothesize that NGF-TrkA signaling endosomes undergo functional diversification allowing for a multitude of outputs, such as survival and synapse development. This diversification can be achieved through unique trafficking steps and association with distinct effector proteins. Our lab has previously identified the first known effector of the retrograde signaling endosome in the soma, Coronin-1a. Loss of Coronin-1a causes a decrease in the number of surviving neurons in vivo, fewer long lived signaling endosomes, and loss of a new trafficking event that we call signaling transcytosis. We have identified a second novel effector of the retrograde signaling endosome in the soma: Neuron enriched endosomal protein of 21kDa (Neep21). In the CNS, Neep21 has been shown to affect the trafficking of multiple receptors, including promoting transcytosis and lysosomal evasion. However, it's role in the PNS is unknown. We show that Neep21 is present both in tissues dependent on NGF-TrkA for survival and on TrkA signaling endosomes. In regard to the trafficking of retrograde TrkA, Neep21 is present on the signaling endosome but does not co-localize with Coronin-1a. In contrast to knockout of Coronin-1a, we show that Neep21 knockout causes re-routing of signaling endosomes through the signaling transcytosis pathway. These data suggest a model of molecular diversification of the TrkA signaling endosome in order to achieve functional specificity with Coronin-1a and Neep21 playing distinct roles.

Disclosures: K.A. Barford: None. **K. McDaniel:** None. **C. Yap:** None. **C. Deppmann:** None. **B.R. Winckler:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.22/B23

Topic: A.05. Axon and Dendrite Development

Support: GRF 15326416

PolyU internal funding 1-ZVGK

PolyU internal funding 1-YW0Q

Title: Ultrasound modulate on axonal function

Authors: *R. R. ZHANG1, Z. QIU, S. KALA, L. SUN
The Interdisciplinary Div. of Biomed. Engin., The Hong Kong Polytechnic Univ., Hong Kong, Hong Kong

Abstract: In central nerve system, each neuron generates forces to extend an axon during development and to perform axonal function such as the formation of dendritic spines during learning and memory. In addition, the neurons are subjected to heterogeneous mechanical environment. How the mechanical properties of CNS influence axon growth and axon functioning in vivo are currently unknown, and the potential neuronal response to mechanical signals in vivo is poorly understood. To date, it is emergent to develop a mechanical based stimulation strategy for in vivo studies. Ultrasound is considered as a promising tool for brain stimulation. It is shown to be able to trigger neuron like cells to outgrowth, axon branching, and accelerate nerve regeneration. We hypothesize that neurons can sense ultrasonic waves by mechanosensitive ion channels e.g. piezo1, to regulate the axon development and functioning. To test this hypothesis, N2A, retinal ganglion cell (RGC), and primary cultured neurons, with high Piezo1 expression, were utilized as in vitro model. Piezo1 knockdown cells with siRNA and Piezo1 blocker GsMTx-4 pre-treated cells were severed as control to test the role of Piezo1 in sensing ultrasound. The real-time calcium imaging was used to test the acute effects of ultrasound on neurons and axons. The axon lengths and structure after 24 h, the extension velocity of axons for all the experimental groups were analyzed from live cell imaging data. In addition, the dynamics of actin and tubulin were visualized in live neurons. At last, the expression level and activity of CREB which are correlated to learning and memory were also investigated by western blot and fluorescence energy transfer imaging (FRET). Our results show that ultrasound stimulation on wild type N2A cell and primary cultured RGC could induce calcium signaling on cell body and axons and promote axon outgrowth. The long term stimulation on primary neurons could increase the generation of dendritic spines. Ultrasound stimulation could modulate the actin and tubulin dynamics and activate CREB activity. The axon length and the extension velocity for Piezo1 knockdown and GsMTx-4 blocked neurons is significant decreased. These results showed that ultrasound as a mechanical wave is able to stimulate neurons and modulate the axon function. The detailed biophysical and molecular mechanism remains unclear the role of piezo1 needs to be confirmed. Given the ability of focusing into small region in deep human brain non-invasively, it is capable to investigate the mechanisms of mechanical effects on neural axon function in vivo.

Disclosures: R.R. Zhang1: None. **Z. Qiu:** None. **S. Kala:** None. **L. Sun:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.23/B24

Topic: A.05. Axon and Dendrite Development

Support: NIH Grant 1R15AG045820-01A1

Summer Research Award from George Mason University

Title: Neuromorphological characterization of CA1 pyramidal cells expressing chimeric NMDAR GluN2 subunits: Changes during hippocampal development

Authors: *C. HUNG[1], M. J. KEITH[2], R. E. KEITH[3], M. F. BADAKHSH[4], T. C. DUMAS[5]
[1]Krasnow Inst. For Advanced Study, Fairfax, VA; [2]Northern Virginia Community Col., Fairfax Station, VA; [3]Neurosci. Dept., [4]Neurosci., [5]Psychology, George Mason Univ. Krasnow Inst. for Advanced Study, Fairfax, VA

Abstract: N-methyl-D-aspartate receptors (NMDARs) at hippocampal excitatory synapses are largely responsible for dendritic and synaptic development. Two signaling properties of NMDARs that have been independently linked to hippocampal plasticity are calcium conductance into the postsynaptic spine and direct intracellular protein interactions. These properties vary with the composition of the NMDAR such that, compared to NMDARs with GluN2A subunits, NMDARs with GluN2B subunits conduct calcium for a longer period following activation and display greater affinity for the synaptic plasticity protein, CaMKII. Conductance regulating domains exist in the GluN2 extracellular amino (A) terminus and transmembrane (TM) regions. Intracellular signaling domains exist in the intracellular carboxy (C) terminus. Prior to the third postnatal week, most NMDARs contain GluN2B. Across the third postnatal week, GluN2B subunits are replaced by GluN2A subunits. To determine the separate influences of the ionotropic and metabotropic properties of NMDARs on dendritic development, we constructed GluN2 chimeras and generated two transgenic mouse lines, one expressing the A-terminus and TM regions of GluN2A fused to C-terminus of GluN2B (termed ABc) and, vice versa, the other expressing the A-terminus and TM regions of GluN2B fused to C-terminus of GluN2A (termed BAc). Transcription was regulated by the TET-off expression system with tetracycline transactivator protein (tTA) expression under control of the CaMKII minimal promoter. tTA expression was seen in many forebrain regions, but predominantly in hippocampal pyramidal cells. We measured neuromorphological characteristics of hippocampal CA1 pyramidal neurons in two distinct postnatal periods: P17-P19, and P22-P24. Utilizing Thy-1 GFP fluorescence methods, confocal microscopy, and Neurolucida tracing, dendritic arbor complexity was measured. Sholl analyses demonstrate a trend for increased arbor length in ABc animals and a smaller arbor in BAc animals at P17-P19. This is also true of the basal dendrite, with neurons in ABc mice exhibiting higher length and branch intersections. Branching analyses results agree with these trends. In the P22-P24 cohort, there are fewer differences between genotypes. While these results are not yet based on comparative statistics, and current sampling is low, these preliminary results suggest that GluN2B C-terminal signaling in ABc animals increases dendritic growth, while in BAc animals, the GluN2A C-terminus reduces dendritic arbor size. Overall, these results support the need for further dissection of GluN2 subunit signaling domains with respect to hippocampal maturation.

Disclosures: C. Hung: None. **M.J. Keith:** None. **R.E. Keith:** None. **M.F. Badakhsh:** None. **T.C. Dumas:** None.

Poster

367. Dendrite Growth and Branching

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 367.24/B25

Topic: A.05. Axon and Dendrite Development

Title: Single cell analysis of Purkinje cell death induced by diphtheria toxin

Authors: T. R. IQBAL, *L. MA
Neurosci., Thomas Jefferson Univ., Philadelphia, PA

Abstract: Cell ablation at the single cell level allows us to address how an individual cell responds to direct injury and how neighboring cells respond to cell death. Here, we study cell death in Purkinje cells (PC) using Cre-induced expression of Diphtheria toxin receptor (DTR) in transgenic mice. Cre delivered specifically to PCs via adeno-associated virus-8 (AAV8) renders only a few cells sensitive to diphtheria toxin (DT), making it feasible to study single cell responses. AAV8 that expresses Cre and green fluorescent protein (Cre-GFP) is co-injected with AAV8 expressing red fluorescent protein (RFP) into the cerebellum of DTR mice at p0-3. While Cre-GFP-PCs express DTR, RFP-PCs do not and thus serve as a control. To induce cell death, DTR mice were injected intraperitoneally with DT at postnatal day (P) 21. An age matched group was injected with saline as a control. Cerebella were collected 1-3 days later for morphological analysis. First, DTR homozygous mice were given a single dose of DT at 100 or 400 ng. Analysis of the ratio of GFP and RFP cells (1:1 GFP:RFP) showed no significant change between single DT application and saline controls. Next, homozygous DTR mice were given DT at 100 or 400 ng once a day for 5 consecutive days. This treatment revealed a significant decrease in GFP cells compared to RFP cells when compared to saline controls (from 2:1 to 1:1 GFP:RFP), demonstrating that GFP-expressing PCs are susceptible to DT. Next, in order to test the effect of gene dosage, heterozygous DTR mice were treated with 100 ng DT for 5 days. The decrease in GFP cells was less pronounced (from 6:1 to 5:1 GFP:RFP), suggesting the dosage dependence of DT-induced PC death. Under these conditions, there were no obvious cellular changes in an intermediate stage of degeneration, that is, membrane blebbing or reduced branching. Finally, in an effort to capture these cellular changes, DTR homozygous and DTR heterozygous mice were treated with DT for 3 days, and analyzed 1 or 3 days after treatment. Preliminary studies revealed some examples of changes in dendritic trees, specifically loss of self-avoidance. Thus, application timing and changes to DTR levels can allow different stages of cell death to be visualized. Further development of these DT-directed ablation paradigms can be used to characterize aspects of cellular degeneration in the cerebellum. For example, determining whether tiling is altered or maintained after death of neighboring cells, or whether dendrites or axons are more susceptible to damage.

Disclosures: **T.R. Iqbal:** None. **L. Ma:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.01/B26

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: ANR-14-CE13-0032-01

Title: Caffeine exposure during hippocampal synaptogenesis alters network circuitry and hippocampal-dependent memory

Authors: ***S. LÉVI**, J. C. PRESSEY, F. G. CASTRO, M. GOUTIERRE, M. RUSSEAU, J. PONCER
INSERM, Paris, France

Abstract: In the adult brain, adenosine, a degradation product of ATP, controls neurotransmitter release and synaptic plasticity through G protein coupled A1 and A2A receptors. The activity of these receptors is essential in normal behaviour, including learning and memory, sleep and arousal, locomotor activity and exploration, feeding behaviour and mood and motivation. However, the role of these receptors in development is not well known. In collaboration with C. Bernard's laboratory (INS, Marseille), we have recently identified a novel mechanism by which the adenosine signaling pathway acts as a detector and stabilizer of active nascent inhibitory GABAergic synapses in primary hippocampal cultures. Based on recent findings, we propose that the deleterious consequences of *in utero* and post-natal brain exposure to caffeine (Sci Transl Med. 2013; 5(197), an antagonist of A1 and A2A receptors, are primarily due to A2A receptor-dependent alterations in synaptogenesis. We are now testing this hypothesis *in vivo* by assessing synaptic protein expression, synapse density as well as neuronal and network activity in the hippocampus. These experiments are performed in both juvenile and adult mice injected daily during the period of hippocampal synaptogenesis (post-natal days 3-16) with caffeine or the selective A2A receptor antagonist SCH58261. Supporting our hypothesis, our data demonstrate caffeine and SCH85261 treatments alter hippocampal synaptogenesis, increase network activity, and leads to long-term deficits in hippocampal-dependent memory tasks in adult mice. Furthermore, juvenile animals exhibited sensitivity to pharmacologically induced epilepsy after treatment with caffeine and SCH85261. Our findings provide a better understanding of the pathological mechanisms engaged upon early-life exposure to caffeine.

Disclosures: **S. Lévi:** None. **J.C. Pressey:** None. **F.G. Castro:** None. **M. Goutierre:** None. **M. Russeau:** None. **J. Poncer:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.02/B27

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: AXA Research Fund

ANR ADONIS

Title: Adenosine A_{2A} receptor: A molecular switch to stabilize/eliminate inhibitory synapses during development

Authors: *S. ZAPPETTINI[1], *S. ZAPPETTINI[1], F. GOMEZ-CASTRO[2,3,4], C. G. SILVA[5], J. C. PRESSEY[2,3,4], M. RUSSEAU[2,3,4], E. EUGÈNE[2,3,4], P. M. CANAS[5], F. Q. GONÇALVES[5], S. ALÇADA-MORAIS[5], E. SZABÓ[5], R. J. RODRIGUES[5], P. AGOSTINHO[5,6], A. R. TOMÉ[5], C. LETTERRIER[7], B. TESSIER[8], B. DARGENT[7], S. K. TYAGARAJAN[9], O. THOUMINE[8], R. A. CUNHA[5,6], M. ESCLAPEZ[1], S. LÉVI[2,3,4], C. BERNARD[1]
[1]INSERM U1106 INS, Marseille, France; [2]INSERM UMR-S 839, Paris, France; [3]Univ. Pierre et Marie Curie, Paris, France; [4]Inst. du Fer a Moulin, Paris, France; [5]CNC-Center for Neurosci. and Cell Biology, Univ. of Coimbra, Coimbra, Portugal; [6]Inst. of Biochemistry, Fac. of Medicine, Univ. of Coimbra, Coimbra, Portugal; [7]Axonal Domains Architecture, Aix Marseille Université, CNRS, CRN2M UMR7286, Marseille, France; [8]Interdisciplinary Inst. for Neuroscience, UMR CNRS 5297- Univ. of Bordeaux, Bordeaux, France; [9]Inst. of Pharmacol. and Toxicology, Univ. of Zürich, Zurich, Swaziland

Abstract: In the adult brain, adenosine, controls neurotransmitter release mainly through inhibitory A1 and facilitator A2A receptors. However, its role in development remains to be elucidated. Here, we addressed the role of A2AR-mediated signalling during GABAergic synaptogenesis in the hippocampus. We found a preferential postsynaptic expression of A2ARs during synaptogenesis in the hippocampus in vitro and in vivo. This developmental expression of A2ARs was correlated with a role of A2ARs in the stabilization of nascent GABA synapses during synaptogenesis. Downregulating A2AR expression with a shRNA approach in isolated postsynaptic cells led to a loss of synapses equivalent to that seen following A2AR activity blockade, demonstrating that the A2AR-mediated synapse stabilization is a cell autonomous process that requires A2AR activation in the postsynaptic cell. ATP and adenosine can be secreted by both glia and neurons. We found that activity-dependent release of neuronal ATP/adenosine is sufficient to stabilize newly formed GABA synapses. Using live cell imaging, we showed that adenosine signalling stabilizes active nascent inhibitory synapses. We then characterized the molecular mechanism downstream postsynaptic A2AR. We identified the Adenylyl cyclase/cAMP/Protein Kinase A (PKA) signalling cascade as the main molecular

pathway and we identified the postsynaptic scaffolding molecule gephyrin as the main target of PKA activation. Finally, we showed the A2AR-mediated stabilization of the post- and pre-synapse required the trans-synaptic Slitrk3-PTPδ complex. These results allow us to propose that A2ARs act as coincidence detectors to stabilize newly formed GABAergic synapses during synaptogenesis.

Disclosures: S. Zappettini: None. **F. Gomez-Castro:** None. **C.G. Silva:** None. **J.C. Pressey:** None. **M. Russeau:** None. **E. Eugène:** None. **P.M. Canas:** None. **F.Q. Gonçalves:** None. **S. Alçada-Morais:** None. **E. Szabó:** None. **R.J. Rodrigues:** None. **P. Agostinho:** None. **A.R. Tomé:** None. **C. Letterrier:** None. **B. Tessier:** None. **B. Dargent:** None. **S.K. Tyagarajan:** None. **O. Thoumine:** None. **R.A. Cunha:** None. **M. Esclapez:** None. **S. Lévi:** None. **C. Bernard:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.03/B28

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: The Ministry of Education, Science, and Culture

Title: Functional mapping of neuronal activity in the facial nucleus of the rat embryo: Optical recording with a voltage-sensitive dye

Authors: *K. SATO[1], Y. MOMOSE-SATO[2]
[1]Dept. of Hlth. and Nutr. Sci., Komazawa Women's Univ, Fac. of Human Hlth., Tokyo, Japan; [2]Dept. of Nutr. and Dietetics, Kanto Gakuin University, Col. of Nutr., Yokohama, Japan

Abstract: During mammalian development, facial motoneurons are generated in rhombomere 4 and migrate caudally, forming a unique axonal trajectory called the "genu", a loop of facial motor axons around the abducence nucleus. It is one of the interesting questions in developmental neuroscience when and how this unique structure is established, and what the correlation between morphogenesis and functiogenesis is. We applied optical recording with a voltage-sensitive dye (VSD) and the DiI staining method to the facial nerve (N.VII)-brainstem preparations dissected from E16-18 rat fetuses, and examined developmental processes of the motor nucleus/bundles of the N.VII. The DiI staining revealed the location of the facial motor nucleus and the pathway of the N.VII. The VSD recording showed that (1) two types of fast spike-like signals, which corresponded to action potentials, were detected, and (2) the fast signal identified in the facial motor nucleus exhibited a long duration, whereas those detected in the N.VII bundles had a shorter duration. We suceeded in analyzing spatiotemporal patterns of N.VII

responses in the early developing rat brainstem, which confirmed the usefulness of VSD recording in funcional mapping of a 3D-complexed neuronal population.

Disclosures: K. Sato: None. **Y. Momose-Sato:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.04/B29

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: The Ministry of Education, Science, and Culture, Japan

The Japan Epilepsy Research Foundation

Title: In ovo blockade of the spontaneous depolarization wave inhibits the formation of synaptic networks in the embryonic brainstem

Authors: *Y. MOMOSE-SATO[1], K. SATO[2]
[1]Dept. of Nutr. and Dietetics, Kanto Gakuin University, Col. of Nutr., Yokohama, Japan; [2]Dept. of Hlth. and Nutr. Sci., Komazawa Women's Univ, Fac. of Human Hlth., Tokyo, Japan

Abstract: During development, spontaneous activity is observed well before sensory pathways are functionally organized. One of the earliest activities expressed within the central nervous system is a widely propagating wave-like activity, which we referred to as the depolarization wave. The depolarization wave is expressed during a specific period of embryogenesis, at which time functional synapses are generated between cranial/spinal sensory nerves and postsynaptic neurons in the brain/spinal cord. This developmental profile has led to the hypothesis that the depolarization wave plays a fundamental role in synaptogenesis, which is known to be activity-dependent. In the present study, we tested this hypothesis by blocking the depolarization wave *in ovo* and examining its effects on functional synaptogenesis in vagus nerve-related brainstem nuclei. Postsynaptic responses were detected from the nucleus of the tractus solitarius (NTS), the contralateral non-NTS region, and the parabrachial nucleus (PBN) using optical recordings with a voltage-sensitive dye. Chronic inhibition of the depolarization wave had no significant affect on the developmental time course, amplitude, and spatial distribution of monosynaptic EPSPs (excitatory postsynaptic potentials) in the first-order nuclei of the vagal sensory pathway (the NTS and the contralateral non-NTS region), but markedly reduced polysynaptic responses in the higher-order nucleus (the PBN). These results suggest that the depolarization wave plays an indispensable role in the initial process of functional synaptic expression in the brainstem, especially in the higher-order nucleus of the cranial sensory pathway.

Disclosures: Y. Momose-Sato: None. **K. Sato:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.05/B30

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Title: Molecular dissection of Neuroligin2 and Slitrk3 reveals an essential framework for GABAergic synapse development

Authors: *J. LI[1], W. HAN[1], X. MAO[1], Y.-X. WANG[2], L. DONG[3], R. S. PETRALIA[2], W. LU[1]
[1]Lab. of Synapse and Neural Circuit, NINDS, NIH, Bethesda, MD; [2]Advanced Imaging Core, NIDCD/NIH, Bethesda, MD; [3]Genet. Engin. Core, NEI/NIH, Bethesda, MD

Abstract: In the mammalian central nervous system, GABAergic interneurons provide the majority of inhibitory synaptic transmission onto principal neurons to balance glutamatergic excitation. Imbalance of excitation and inhibition in synaptic transmission has been implicated in many psychiatric and neurological disorders, such as epilepsy, schizophrenia, and autism spectrum disorders (ASD). Thus, it is important to understand the molecular mechanisms underlying the development of excitatory and inhibitory synapses. While extensive studies have been focused on the development of glutamatergic excitatory synapses, much less is known about inhibitory synaptogenesis. Recently, numerous molecules, including trans-synaptic adhesion molecules, sub-membranous scaffolding proteins, intracellular signaling molecules, and secreted factors, have been identified to function in inhibitory synapse development. However, it remains largely unknown whether there is a unifying mechanism for development of diverse inhibitory synapses. In the present study, we have employed cell biological and biochemical techniques, and shRNA knockdown, molecular replacement and gene targeting approaches to examine two inhibitory synaptic cell adhesion molecules (CAMs), Neuroligin2 (NL2) and Slitrk3 (ST3), in GABAergic synapse development in hippocampal neurons. We have found that, in the early developing neurons, the establishment of GABAergic synapses mainly depends on NL2, but not ST3. As the neuron matures, both NL2 and ST3 are required for GABAergic synaptogenesis. Importantly, NL2 and ST3 interact with each other through their extracellular domains. Functionally, NL2 interaction with ST3 facilitates ST3 trafficking to the plasma membrane and augments their synaptogenic ability to induce GABAergic synapse formation. Furthermore, perturbation of the NL2-ST3 interaction strongly impairs inhibitory synaptic transmission *in vitro*. We have also generated mutant mouse line in which the NL2-ST3 interaction is abolished, and have found that both GABAergic synaptic transmission and synapse density in hippocampal CA1 neurons are significantly reduced, suggesting that the NL2-ST3 interaction is critical for GABAergic synapse development *in vivo*. In summary, these findings demonstrate how different postsynaptic CAMs, NL2 and ST3, work in concert to control

inhibitory synaptogenesis, and establish a general molecular framework essential for GABAergic synapse development in hippocampus.

Disclosures: **J. Li:** None. **W. Han:** None. **X. Mao:** None. **Y. Wang:** None. **L. Dong:** None. **R.S. Petralia:** None. **W. Lu:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.06/B31

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: GM108539

NS081707

Title: Trans-synaptic regulation of sensory circuit connectivity in the spinal cord by neurexins and neuroligins

Authors: *B. A. COPITS, K. C. MCKENZIE, J. J. YOO, S. K. VOGT, R. W. GEREAU, IV
Pain Center, Dept of Anesthesiol., Washington Univ. Sch. of Med., Saint Louis, MO

Abstract: Understanding the assembly and wiring of neural circuits, and how they integrate and process information, is a formidable task. Developmental studies at the interface of the central and peripheral nervous systems have provided significant insights into how diverse molecular families coordinate the positioning and innervation of target regions in the spinal cord. However, we lack a detailed understanding of how connectivity in these circuits is specified. Synaptic adhesion molecules are key coordinators of synapse specificity and diversity, and *trans*-synaptic interactions between the large variety of alternatively spliced presynaptic neurexins and postsynaptic neuroligins have been suggested to impart a combinatorial code for neural connectivity. Despite this hypothesis, little is known about how these molecules govern synapse and circuit formation to specify behavioral responses.
Somatosensory circuits represent intriguing systems for studying the molecular and cellular mechanisms of synapse specificity and connectivity. The wealth of unique recombinase-expressing mouse lines permit genetic access to molecularly-defined populations of sensory neurons without confounding effects in other regions of the nervous system. Additionally, there exists a substantial number of well-defined modality-specific behavioral tests to investigate how distinct molecular families alter circuit processing. Here we employ a conditional genetic strategy to understand how presynaptic neurexin ligands specify synaptic connectivity of sensory circuits in the spinal cord. We found that genetic deletion of β-neurexins in $Na_v1.8+$ sensory neurons, which includes both nociceptors and mechanosensitive fibers, led to modality specific

alterations in sensory behaviors. While these manipulations did not alter mechanical or heat sensitivity, mice exhibited an increased sensitivity to cold. Despite normal heat thresholds, inflammation-induced thermal hypersensitivity, which engages plasticity mechanisms in the spinal cord, was completely absent. To understand how these manipulations regulate synaptic plasticity and connectivity, we have created conditional optogenetic knockout lines to selectively stimulate fibers lacking neurexins. In spinal cord slices, loss of neurexins weakens synaptic transmission, alters AMPA/NMDAR ratios, and decreases synaptic connectivity. We are now using a combination of approaches, both *in vivo* and *in vitro*, to understand how distinct combinations of neurexins and neuroligins regulate synaptic specificity in these circuits.

Disclosures: B.A. Copits: None. **K.C. McKenzie:** None. **J.J. Yoo:** None. **S.K. Vogt:** None. **R.W. Gereau:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.07/B32

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: NIH NINDS (R01 NS072129)

NSF (IOS-1121095)

Title: A MIG-15/JNK-1 MAP kinase cascade opposes RPM-1 signaling in synapse formation and learning

Authors: *B. GRILL, A. C. GILES, R. BIRNBAUM, S. KASHYAP, O. CRAWLEY
Neurosci., Scripps Res. Inst., Jupiter, FL

Abstract: The Pam/Highwire/RPM-1 (PHR) proteins are conserved intracellular signaling hubs that regulate synapse formation and axon termination. The *C. elegans* PHR protein, called RPM-1, acts as a ubiquitin ligase to inhibit the DLK-1 and MLK-1 MAP kinase pathways. We have identified a new MAP kinase pathway that suppresses synapse formation defects, but not axon termination defects, in the mechanosensory neurons of *rpm-1* mutants. This pathway includes: MIG-15 (MAP4K), NSY-1 (MAP3K), JKK-1 (MAP2K) and JNK-1 (MAPK). Transgenic overexpression of kinases in the MIG-15/JNK-1 pathway is sufficient to impair synapse formation in wild-type animals. The MIG-15/JNK-1 pathway functions cell autonomously in the mechanosensory neurons, and kinases in the pathway localize to presynaptic terminals providing further evidence of a role in synapse development. Behavioral analysis indicates loss of MIG-15/JNK-1 signaling increases habituation to repeated mechanical stimuli, a type of short-term learning that is dependent upon mechanosensory neurons. Habituation results also show the

MIG-15/JNK-1 pathway functions as a parallel opposing pathway to RPM-1. Thus, the MIG-15/JNK-1 pathway is required to restrict both glutamatergic synapse formation and short term learning.

Disclosures: B. Grill: None. **A.C. Giles:** None. **R. Birnbaum:** None. **S. Kashyap:** None. **O. Crawley:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.08/B33

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: NIH Grant T32 NS061764

NIH Grant DA018928

Title: Roles of the Rac1 activator Farp1 in mammalian synapse and dendrite development

Authors: *A. J. COLEMAN[1], L. LI[2], J. COTTRELL[2], T. BIEDERER[3]
[1]Neurosci. Grad. Program, Tufts Univ. - Sackler Sch., Boston, MA; [2]Stanley Ctr. for Psychiatric Res., Broad Inst. of MIT and Harvard, Cambridge, MA; [3]Dpt. of Neurosci., Tufts Univ. Sch. of Med., Boston, MA

Abstract: Regulators of the actin cytoskeleton are critical for the structural and functional maturation of neurons, and mutations in genes encoding regulators of the cytoskeleton are linked to intellectual disability and autism-spectrum disorders. Select small G proteins, including the Rho GTPase Rac1, control actin dynamics in developing neurons and at excitatory synapses. The differential roles of Rac1 regulators across brain regions and during specific developmental time windows remain an important knowledge gap. We have previously characterized the roles of the Rac1 activator Farp1 (FERM RhoGEF and pleckstrin domain-containing protein 1) in regulating synapse and dendrite development in hippocampal neurons. We report here that an autism-linked variant of Farp1 exhibits reduced expression. To address the roles of Farp1 and investigate loss-of-function effects, we have generated Farp1 conditional knockout (cKO) mice. Glutamatergic neurons from juvenile Farp1 cKO mice exhibit altered dendrite structure, including reduced spine density in CA1 pyramidal neurons. We have probed the effect of Farp1 loss on its signaling partners including the trans-synaptic adhesion molecule SynCAM1 and members of the Semaphorin/Plexin signaling pathway. Additionally, we have characterized the role of Farp1 in hippocampal-dependent learning and memory in juvenile and adult animals. These *in vivo* studies are complemented by an *in vitro* analysis of dynamic roles of Farp1 in modulating synaptic plasticity and we report on the contribution of activity-dependent phosphorylation of

Farp1 to synaptic remodeling in long-term potentiation. Our results highlight the functions of Farp1 in neuronal differentiation and how its activity is modulated by its signaling partners to regulate multiple aspects of connectivity.

Disclosures: A.J. Coleman: None. **L. Li:** None. **J. Cottrell:** None. **T. Biederer:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.09/B34

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: Swedish Research Council 350-2012-6543

NIMH Grant MH052804

NIMH Grant MH104172

Title: The carbonic anhydrase-related proteins ca10 and ca11 are conserved pan-neurexin cis-ligands

Authors: *F. H. STERKY[1], J. H. TROTTER[2], S. LEE[3], T. C. SUDHOF[2]
[1]Clin. chemistry and transfusion medicine, Univ. of Gothenburg, Gothenburg, Sweden; [3]Mol. and Cellualr physiology, [2]Stanford Univ., Stanford, CA

Abstract: Neurexins are pre-synaptic cell adhesion receptors that interact with diverse post-synaptic ligands to shape the molecular architecture and properties of the synapse. Mutations in neurexin-encoding genes have repeatedly been linked to neuropsychiatric disease, but despite being extensively studied, the physiological functions of the neurexin complex and role in human disease are not well understood. In proteomic studies of Nrxn1 complexes immuno-isolated from mouse brain, we identified carbonic anhydrase-related proteins CA10 and CA11, two homologous, secreted glycoproteins of unknown function that are predominantly expressed in brain. We found that CA10 directly binds in a *cis*-configuration to a conserved membrane-proximal, extracellular sequence present in □α-, β- and γ-neurexins. The CA10/neurexin complex is stable and stoichiometric, and results in formation of an intermolecular disulfide bond between conserved cysteine residues in neurexins and CA10. This interaction promotes surface-expression of α- and β-neurexins, suggesting that CA10 may form a complex with neurexins in the secretory pathway that facilitates surface-transport and/or stability of neurexins. These results expand our understanding of the neurexin complex and provide a means to further address its posttranslational regulation.

Disclosures: F.H. Sterky: None. **J.H. Trotter:** None. **S. Lee:** None. **T.C. Sudhof:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.10/B35

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: HD001205 -23 DIR

Title: Lissencephaly heterotopia and effects on the development of cellular microcircuitry

Authors: *J. D'AMOUR[1], T. G. EKINS[3], C. J. MCBAIN[2]
[2]Lab. Cell/Molec Neurosci, [1]NIH, Bethesda, MD; [3]NIH/Brown Univ., Bethesda, MD

Abstract: In this study, we are examining cellular layering and its role in the development of synaptic microcircuitry. Principal cells of the developing mouse hippocampus arise in proliferative waves of cell birth and subsequent radial migration over embryonic day 13-16, to ultimately appear as a compacted layer of excitatory neurons. In the neocortex a similar process of cell birth but temporally separate radial migration patterns produces six distinct cell-layers in an inside-out fashion. The resulting cortical layers have specialized microcircuitry essential to information processing carried out in that cortical region. Here we investigate whether similar excitatory cell sub-populations and birthdate specified microcircuitry also exist within the hippocampal principal cell layer (PCL). Among the first evidence of excitatory subpopulations in the radial axis of the PCL (superficial to deep) in region CA1, is restricted calbindin expression to the superficial rows of principal cells, while deeper principal cells are typically calbindin negative. Recent studies further support radial subdivisions among PCL excitatory cells and suggest an intricate synaptic connectivity of local excitatory and inhibitory sub-populations. However, it remains a daunting problem to disentangle the effects of cellular birthdate, positioning and genetic identity in the establishment of microcircuitry. Toward this goal, we make use of a mouse model of a human neurodevelopmental disorder, in which cellular layering and positioning are effected. Lissencephaly occurs in about 0.001% live human births, and is caused by mutations in genes essential to cell migration. In hippocampi from heterozygous Lis1 mice (Lis+/-), the PCL is often fragmented into radially divided heterotopic bands or cell-clusters. Our data from Lis+/- animals indicate that calbindin positive principal cells occupy the deeper heterotopic cell band (in contrast to their superficial positioning in the wild type PCL) and may reflect an inversion of excitatory cell positioning. This observation could carry interesting ramifications for inhibitory connectivity, particularly among parvalbumin positive interneurons that have been demonstrated to preferentially innervate deeper principal cells in the wild-type hippocampus, area CA1. Additionally, our parvalbumin and somatostatin staining indicate these interneuron types migrate to abnormally superficial locations. Future experiments

will examine connectivity between these cell types, with hopes of better understanding how developmental challenges such as cellular heterotopia shape synaptic connectivity.

Disclosures: **J. D'Amour:** None. **T.G. Ekins:** None. **C.J. McBain:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.11/B36

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: NIH Grant F31 DC014393

NIH Grant R01 DC007695

NIH Grant F32 DC008730

NIH/NIGMS CoBRE Grant P20 GM103503

NIH/NIGMS Grant P41 GM103412

Title: A novel cell polarity program during innervation of a non-laminar nucleus in the auditory brainstem

Authors: ***P. S. HOLCOMB**[1], A. N. BRANDEBURA[1], D. R. KOLSON[1], S. H. BERZINGI[1], D. R. JACKSON[1], T. J. DEERINCK[2], M. H. ELLISMAN[2], G. A. SPIROU[1]
[1]Sensory Neurosci. Res. Ctr., West Virginia Univ. Sch. of Med., Morgantown, WV; [2]Natl. Ctr. for Microscopy and Imaging Res., Univ. of California San Diego, La Jolla, CA

Abstract: Laminar sensory structures—rod and cone cells of the retina, hair cells of the cochlea—confine innervation to specific sites through polarized expression of synaptic proteins and lateral cell-cell connections that structurally segregate these cells into apical and basal regions. Many cells in the central nervous system, however, are not arranged in a laminar fashion necessary to make such a polarized distinction. Are these populations of cells still structurally polarized, and does this polarity inform the placement and growth of synaptic terminals? The calyx of Held (CH) - principal cell (PC) connection in the medial nucleus of the trapezoid body (MNTB) of the mouse provides a valuable model system to examine these questions, due to its short developmental window (<72 hours), large terminal size, and a clear monoinnervated endpoint. We acquired, segmented, and reconstructed PCs and their associated terminals from postnatal day (P)2-9 mice using serial block-face scanning electron microscopy and both manual and semi-automated segmentation techniques. An “intrasomatic polarity”, or asymmetric arrangement of the nucleus and organelles into a nucleus-poor “cytoplasmic” pole and a nucleus-

filled “nuclear” pole, was present in all PCs reconstructed (n=97) This polarity increases significantly during development due to asymmetric growth of the PC away from the nucleus, and relaxes somewhat by P30, suggesting a role specifically in development. Additionally, the nucleus is morphologically polarized, with large invaginations solely on the surface facing the cytoplasmic pole. Polarity also influenced the location of the growing calyx; we found that the CH:PC connection not only becomes larger but also more polarized as development progresses, with greater than 80% of the ASA of the largest terminals at P6 occupying the cytoplasmic pole surface opposite the nucleus. Cells with competing inputs (2-3 inputs/cell with less than 1:5 ratio in size) fell into three categories: (1) the largest terminal is most polarized, (2) the second largest terminal is most polarized, or (3) the two largest terminals are equally polarized. This suggests a “flip-flop” of largest for second largest terminal may be taking place, such as is seen in neuromuscular junction development. These findings demonstrate a novel cell polarity program in a non-laminar cell group that influences the placement and growth potential of terminals in the formation of the CH:PC circuit.

Disclosures: P.S. Holcomb: None. **A.N. Brandebura:** None. **D.R. Kolson:** None. **S.H. Berzingi:** None. **D.R. Jackson:** None. **T.J. Deerinck:** None. **M.H. Ellisman:** None. **G.A. Spirou:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.12/B37

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Title: NRX assembles synapses independent of NL and tunes synaptic plasticity by antagonizing NL *In vivo*

Authors: *P. KURSHAN, K. SHEN
Biol. Dept, Stanford Univ., Stanford, CA

Abstract: A central conundrum in the field of synaptic development is why synaptic inducers identified in cell culture assays, such as Neurexin and Neuroligin, fail to show major synaptogenesis defects when knocked out *in vivo*. Instead, their role seems limited to subtle effects on synaptic function and plasticity. Here we report dual, opposing functions of pre- vs post-synaptic Neurexin on both synapse assembly and synaptic plasticity at the same *in vivo* synapse in *C. elegans*. Moreover we report a novel antagonistic relationship between Neurexin and Neuroligin in mediating this plasticity. We find that presynaptic neurexin cell-autonomously mediates synapse assembly, in a manner independent of Neuroligin, and in fact independent of the entire neurexin extracellular binding domain. In contrast, postsynaptic Neurexin at the same synapse induces downregulation of specific active zone proteins involved in presynaptic

plasticity. This inhibitory action of Neurexin is blocked by presynaptic, perisynaptically localized Neuroligin, suggesting a novel antagonistic role of the two proteins in mediating synaptic plasticity.

Disclosures: P. Kurshan: None. **K. Shen:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.13/B38

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: Alzheimer Society Research Program Biomedical Doctoral Awards to Y.N.

Canadian Institutes of Health Research Grant MOP-133517 to H.T.

A Fonds de la recherche du Québec Research Scholars (Junior 2) grant to H.T.

Scottish Rite Charitable Foundation of Canada research grant to H.T.

2016-IRCM-Michel-Bélanger Scholarship to A.K.L.

Title: Inhibitory synapse differentiation promoted by a novel neurexin2α-interacting cell adhesion molecule IgSF21

Authors: *Y. NAITO[1,2], Y. TANABE[1], C. VASUTA[1], A. K. LEE[1,3], Y. SOUMOUNOU[1], M. W. LINHOFF[4,5,6], H. TAKAHASHI[1,7,8]
[1]Synapse Develop. and Plasticity, Inst. de Recherches Cliniques de Montreal, Montreal, QC, Canada; [2]The Integrated Program in Neurosci., Mcgill Univ., Montreal, QC, Canada; [3]Integrated Program in Neurosci., Mcgill, Montreal, QC, Canada; [4]Dept. of Anat. and Neurobio., Washington Univ. Sch. of Med., St. Louis, MO; [5]The Brain Res. Ctr. and Dept. of Psychiatry, Univ. of British Columbia, Vancouver, BC, Canada; [6]Vollum Inst., Oregon Hlth. and Sci. Univ., Portland, OR; [7]Dept. of Med., Univ. de Montréal, Montreal, QC, Canada; [8]Div. of Exptl. Med., McGill Univ., Montreal, QC, Canada

Abstract: Inhibitory synaptic inputs control neuronal excitability and the firing patterns of targeted neurons and further regulate brain circuit formation. The proper balance between excitatory and inhibitory synaptic inputs is crucial for maintaining normal brain functions. Accumulating evidence suggests that impaired development of central inhibitory synapses leads to neuropsychiatric disorders such as schizophrenia, autism spectrum disorders, and anxiety, highlighting the importance of our understanding of the molecular mechanisms of inhibitory synapse development. However, the molecular mechanism governing the development of

GABAergic inhibitory synapses is poorly understood.
Synapse development requires not only physical contact between axons and target neurons but also chemically-matched pre- and post-synaptic differentiation. Synapse organizers, synaptic adhesion molecules with the ability to induce synaptic differentiation, form trans-synaptic complex, called synapse organizing complexes. These complexes have been demonstrated as essential molecular signals for synapse development, represented by the neuroligin (NLG)-neurexin (NRX) complex. Although many synapse organizers induce excitatory or both excitatory and inhibitory synapses, there is only a few synapse organizer selectively inducing inhibitory synapse.
Here, we report the identification of immunoglobulin superfamily member 21 (IgSF21) as a novel inhibitory synapse organizer that induce only inhibitory presynaptic differentiation. Through further proteomics screen, we isolated NRX2α as an IgSF21-interacting presynaptic organizer. Interestingly, IgSF21 selectively binds to NRX2a but not any other NRX isoforms and recruits NRX2α to presynaptic sites in a trans-interaction manner, which is essential for the synaptogenic activity of IgSF21. To characterize the physiological function of IgSF21 in the central nervous system, we comprehensively characterized IgSF21 mutant mice. We found that IgSF21 positively regulates inhibitory presynaptic organization and GABA-mediated synaptic transmission in the hippocampal CA1 pyramidal neurons and that IgSF21 is indispensable for normal sensorimotor gating. Together, our findings suggest that IgSF21 selectively organizes inhibitory synapses via its trans-synaptic interaction with axonal NRX2α and that this is essential for normal brain function.

Disclosures: **Y. Naito:** None. **Y. Tanabe:** None. **C. Vasuta:** None. **A.K. Lee:** None. **Y. Soumounou:** None. **M.W. Linhoff:** None. **H. Takahashi:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.14/B39

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: NUHS seed grant (R-118-00-154-112)

NUHS aspiration grant (R-181-000-164-720)

Title: Expression analysis of miRNA-26b involved in synaptogenesis in embryonic neural stem cells from diabetic mice

Authors: *S. RAMYA[1], S. SHYAMASUNDAR[2], B. BOON HUAT[2], S. T. DHEEN[2]
[1]Anat., Natl. Univ. of Singapore, Singapore, Singapore; [2]Anat., Natl. university of Singapore, Singapore, Singapore

Abstract: Maternal diabetes causes neural tube defects in embryos and neuropsychological deficits in infants. Several molecules and epigenetic factors have been identified to be deregulated in developing brain of embryos by maternal diabetes, although the exact mechanism remains unknown. Recently, miRNAs have been shown to regulate brain development. Therefore, we hypothesized that maternal diabetes alters the expression of miRNAs that regulate genes involved in synaptogenesis and brain development during embryogenesis. In order to address this, embryonic neural stem cells (NSCs) from embryos of normal and streptozotocin-induced diabetic mice were isolated and cultured *in vitro*. High throughput miRNA expression profiling revealed altered expression of several miRNAs in NSCs isolated from embryos of diabetic pregnancy when compared to control. Among the differentially expressed miRNAs, expression of miR-26b, which has been shown to be involved in neurogenesis and neuronal differentiation, was significantly upregulated in NSCs from embryos of diabetic pregnancy when compared to the control. Among the several predicted targets of miR-26b, methyl CpG binding protein2 (Mecp2) and post synaptic density 95 (Psd95) that are involved in brain development, synaptic maturation and plasticity were selected for further study, since the expression levels of Mecp2 and Psd-95 protein were significantly downregulated in NSCs from embryos of diabetic pregnancy when compared to the control. Overexpression of miR-26b resulted in the downregulation of Mecp2 and Psd-95 proteins suggesting that they may be putative targets of miR-26b. These results warrant further investigation as Psd-95 protein is an integral part of the glutamate receptors especially N-methyl-D-aspartate receptor (NMDA) which has a pivotal role in transmission of glutamate in excitatory synapses.
Taken together, this study reveals that maternal diabetes upregulates the expression of miR-26b resulting in down regulation of its predicted targets, Mecp2 and Psd-95 proteins. Decreased expression of Mecp2 and Psd-95 that are crucial for brain development and synapse formation especially excitatory synapses may underlie defective brain development and patterning and manifest as neuropsychological disturbances that are observed in offspring of diabetic pregnancy.

Disclosures: **S. Ramya:** None. **S. Shyamasundar:** None. **B. Boon Huat:** None. **S.T. Dheen:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.15/B40

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: The Royal Society International Exchange Grant IE131294

Title: Investigating the structural role of GABAA receptors in inhibitory synapse formation and circuitry of the basal ganglia

Authors: *J. N. JOVANOVIC[1], J. E. ARAMA[1], S. K. TYAGARAJAN[2], P. PANZANELLI[3], J.-M. FRITSCHY[2]
[1]UCL Sch. of Pharmacy, Univ. Col. London, London, United Kingdom; [2]Univ. Zurich/ Inst. Pharmacol Toxicol, Zurich, Switzerland; [3]Dept. of Neurosci. Rita Levi Montalcini, Univ. of Turin, Turin, Italy

Abstract: Gamma-aminobutyric acid (GABA) is the main inhibitory neurotransmitter in the brain. Its fast synaptic actions are mediated by GABAA receptors (GABAARs), which are chloride/bicarbonate-permeable ligand-gated channels. GABAergic transmission is essential for neuronal development and maturation, and for coordination of neuronal activity underlying all physiological and behavioral processes. The molecules and mechanisms that regulate the assembly and maturation of GABAergic synapses during ontogeny remain largely unknown. Our recent work has demonstrated that GABAARs exhibit synaptogenic activity in heterologous co-culture model systems in which their presence at the postsynaptic plasma membrane is sufficient to promote the adhesion and functional maturation of GABAergic axon terminals. Furthermore, synapse initiation in this system occurs with a high degree of selectivity with respect to the postsynaptic GABAAR composition and it is mediated by the large N-terminal extracellular domains of GABAAR subunits. To investigate this synaptogenic role of GABAARs *in vivo*, we have carried out a quantitative immunohistochemical analysis of GABAergic synapses in the nuclei of the basal ganglia of the alpha1- or alpha2-GABAAR knockout mice using antibodies specific for the main pre- and postsynaptic markers. We have characterized the number and size of the postsynaptic alpha1/2/3 or 5 or gamma2-containing GABAAR clusters, the number of co-localized VGAT or GAD65 positive GABAergic terminals, the number and size of synaptic gephyrin and neuroligin-2 clusters, and the density of TH-positive dopaminergic terminals. Our results demonstrate the loss of inhibitory synapses and profound structural changes in the remaining synapses in those basal ganglia regions in which the most abundantly expressed isoform of the alpha subunits was genetically ablated. This indicates that synaptogenic effects of GABAARs in vivo depend on their individual subunit composition in which the main role in initiation and selectivity of synaptic contacts is mediated by the alpha subunits.

Disclosures: J.N. Jovanovic: None. **J.E. Arama:** None. **S.K. Tyagarajan:** None. **P. Panzanelli:** None. **J. Fritschy:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.16/B41

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: NIH Grant R01MH101102

Title: Prenatal stress induces excitatory-inhibitory imbalance in hippocampus of KCNH2-3.1 transgenic mice

Authors: *Z. HU[1], J. WU[1,2], Y. LI[1,2], M. REN[1], J. L. HILL[1], S. QIN[1], S. ZHU[1,2], Q. TIAN[1], K. MARTINOWICH[1], D. R. WEINBERGER[1], F. YANG[1]
[1]Lieber Inst. for Brain Develop., Baltimore, MD; [2]Col. of Basic Medicine, Chongqing Med. Univ., Chongqing, China

Abstract: The etiology of schizophrenia is largely attributed to genomic variation. However, environmental challenges contribute to the development of schizophrenic symptoms as well. But, the synergistic effect of genetic factors and environmental challenges to schizophrenia remains to be elucidated. Here, by using a genetic animal model of molecular pathology associated with schizophrenia, we tested how life-threatening events during pregnancy affect the pathophysiology of schizophrenia in offspring. KCNH2-3.1, which encodes a primate-specific and brain-selective isoform of the potassium voltage-gated channel, is associated with schizophrenia in gene expression studies in post-mortem brain. We administered a variable and unpredictable stress paradigm to female pregnant mice with transgenic overexpression of human KCNH2-3.1. Prenatal stress resulted in a decrease in mushroom dendritic spine number, as well as spine area, but not in stubby dendritic spine and filopodia, in hippocampal CA1 pyramidal neurons of KCNH2-3.1 transgenic mice at 2 weeks of age compared with control mice at the same age. In addition, we observed a marked increase of inhibitory perisomatic GAD65 synapses on hippocampal CA1 pyramidal neurons in both 2-week-old and 6-week-old KCNH2-3.1 transgenic mice exposed to prenatal stress. Interestingly, this phenotype was not found in GAD67 boutons on CA1 pyramidal cells in KCNH2-3.1 mice after maternal stress. Our results suggest that environmental challenge during pregnancy leads to the imbalance of excitatory-inhibitory synapse formation within the hippocampus in the context of a genetic alteration related to schizophrenia.

Disclosures: Z. Hu: None. **J. Wu:** None. **Y. Li:** None. **M. Ren:** None. **J.L. Hill:** None. **S. Qin:** None. **S. Zhu:** None. **Q. Tian:** None. **K. Martinowich:** None. **D.R. Weinberger:** None. **F. Yang:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.17/B42

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: Damon Runyon Postdoctoral Fellowship

NIH Grant R21 EY025421

Title: Combining RNA-seq and somatic CRISPR mutagenesis to study mouse neural development *In vivo*

Authors: *S. SARIN[1], *. E. ZUNIGA-SANCHEZ[3], Y. KURMANGALIYEV[3], H. COUSINS[2], M. PATEL[2], K. ZHANG[3], M. A. SAMUEL[4], M. MOREY[3], S. ZIPURSKY[3], J. R. SANES[2]
[2]Mol. and Cell. Biol., [1]Harvard Univ., Cambridge, MA; [3]Dept. of Biol. Chem., UCLA, Los Angeles, CA; [4]Dept. of Neuroscience, Huffington Ctr. on Aging, Baylor Col. of Med., Houston, TX

Abstract: Forward genetic screens have been invaluable for identifying genes that regulate synaptogenesis in invertebrates, but few such screens have been performed in mammals owing to the expense and difficulty of germ-line mutagenesis. Here, we propose an alternative approach: purification of synaptic partners at key developmental stages; transcriptional profiling (RNA-seq) to identify candidate mediators of their interactions; and somatic CRISPR/Cas9-based mutagenesis to assess their roles in vivo. We applied this strategy to mouse outer retina, in which axons of rod and cone photoreceptors form synapses on dendrites of rod bipolar and cone bipolar interneurons, respectively, in a thin neuropil called the outer plexiform layer (OPL). We purified and profiled these four cell types, identifying hundreds of genes encoding cell surface and secreted proteins that were differentially expressed among them as synapses are forming. We then generated guide RNAs to inactive a set of them, and introduced them into outer retina by electroporation, along with Cas9 and a fluorescent marker to identify transduced cells. Using this strategy, we found that inactivation of Wnt5a and 5b, which are selectively expressed by rod bipolars, leads to formation of a supernumerary OPL. To investigate Wnt5's mechanism of action, we used cell type-specific promoters to inactivate 8 candidate signal transduction components. We found that Wnt5 acts on rods through a non-canonical pathway using Ryk, Fzd4 and Fzd5 as receptors. Neither the initial screen nor the investigation of signaling mechanism could have been performed in a timely manner using germ-line constitutive and conditional mutagenesis. These methods can be employed to study circuit development in many parts of the developing mouse brain.

Disclosures: S. Sarin: None. ***.E. Zuniga-Sanchez:** None. **Y. Kurmangaliyev:** None. **H. Cousins:** None. **M. Patel:** None. **K. Zhang:** None. **M.A. Samuel:** None. **M. Morey:** None. **S. Zipursky:** None. **J.R. Sanes:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.18/B43

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: VA Merit Review Grant I01-BX002949

VA CDA-2 Grant 005-10S

NIH RO1-NS080979

NIH P30-NS061800

Title: Alpha2delta1-mediated synaptogenesis *In vitro* and during adult neurogenesis

Authors: K. A. BEESON[1], G. L. WESTBROOK[2], *E. SCHNELL[3]
[1]Oregon Hlth. and Sci. Univ., Portland, OR; [2]Vollum Inst., Portland, OR; [3]Portland VA Med. Ctr., Portland, OR

Abstract: Identification of the molecules that control synaptic integration of new neurons is fundamental to our understanding of neural plasticity in health and disease. The α2δ proteins have been identified as receptors for the glial-secreted thrombospondins, which are potent synaptogenic proteins. Excitatory synapse formation is positively regulated by the expression of α2δ during development and in vitro. Although α2δ proteins have been implicated in neural development, and mutations in α2δ can result in striking neurophysiological phenotypes, their contribution to the functional integration of developing and adult-born neurons is not well understood. Interestingly, a commonly prescribed anti-epileptic and analgesic drug, gabapentin, binds α2δ proteins with high affinity and antagonizes thrombospondin-mediated synaptogenic signaling. Using whole cell electrophysiology in organotypic slice cultures, we demonstrate that gabapentin reduces miniature excitatory post-synaptic current frequency, and that α2δ-1 overexpression in single neurons enhances evoked excitatory post-synaptic currents; consistent with a role for α2δ proteins in synaptogenesis. Furthermore, gabapentin significantly reduced dendritic spine width in adult-born granule cells in vivo, indicating that α2δ contributes to synapse formation during adult neurogenesis. Using in vitro and in vivo models of neuronal development paired with single-cell morphology and electrophysiology, we hope to decipher the contribution of α2δ proteins to synaptogenesis in adult-born neurons.

Disclosures: K.A. Beeson: None. **G.L. Westbrook:** None. **E. Schnell:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.19/B44

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: NIH Grant R01 DC-005982

NIH Grant K99 DC-013059

NIH Grant R00 DC-013059

Title: Presynaptic LRP4 promotes synapse number and function of excitatory CNS neurons

Authors: *T. J. MOSCA[1], D. LUGINBUHL[2], I. WANG[3], L. LUO[2]
[1]Neurosci., Thomas Jefferson Univ., Philadelphia, PA; [2]Dept. of Biol., [3]Dept. of Neurobio., Stanford Univ., Stanford, CA

Abstract: Precise coordination of synaptic connections ensures proper information flow within circuits. The activity of presynaptic organizing molecules signaling to downstream pathways is essential for such coordination, though such entities remain incompletely known. We show that LRP4, a conserved transmembrane protein known for its postsynaptic roles, functions presynaptically as an organizing molecule. In the *Drosophila* brain, LRP4 preferentially localizes to excitatory neuron terminals at or near active zones. Loss of presynaptic LRP4 reduces excitatory (not inhibitory) synapse number, impairs active zone architecture, and abolishes olfactory attraction - the latter of which can be suppressed by reducing presynaptic $GABA_B$ receptors. LRP4 overexpression increases synapse number in excitatory and inhibitory neurons, suggesting an instructive role and a common downstream synapse addition pathway. Mechanistically, LRP4 functions via the conserved kinase SRPK79D to ensure normal synapse number and behavior. This highlights a presynaptic function for LRP4, enabling deeper understanding of how synapse organization is coordinated.

Disclosures: T.J. Mosca: None. **D. Luginbuhl:** None. **I. Wang:** None. **L. Luo:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.20/B45

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: NIH Grant R01NS064263 to MMF

NIH Grant F31DA038399 to AP

Title: The synaptic organizer neurexin coordinates cholinergic connectivity with GABAergic neurons

Authors: *M. L. LEMONS[1], A. PHILBROOK[2], S. RAMACHANDRAN[2], D. OLIVER[2], C. LAMBERT[2], M. M. FRANCIS[2]

[1]Dept. of Natural Sci., Assumption Col., Worcester, MA; [2]Neurobio., Univ. of Massachusetts Med. Sch., Worcester, MA

Abstract: In the central nervous system, individual neurons often project divergent synaptic connections to multiple postsynaptic partners. However, we have only a limited understanding of how synaptic specificity across postsynaptic partners is established, and the mechanisms that govern differential patterns of synaptic connectivity. Dyadic excitatory synapses in the *C. elegans* motor circuit provide a useful model for elucidating the molecular basis of synaptic specificity. Excitatory cholinergic motor neurons synapse onto both inhibitory GABAergic neurons and body wall muscles. While genes required for muscle cholinergic synaptic assembly have been well defined, excitatory synaptic connections onto GABAergic neurons appear to require a distinct molecular scaffold. We found that the cell adhesion molecule neurexin/*nrx-1* is specifically required for the assembly of synaptic inputs onto GABAergic neurons. In the absence of *nrx-1*, both cholinergic receptor localization and the formation of synapse-associated morphological features of GABAergic dendrites are disrupted. As a result, excitatory transmission onto GABAergic neurons is severely impaired. *nrx-1* expression in cholinergic neurons is coregulated with cholinergic neuron identity, and is required for proper synaptic connectivity with GABAergic neurons. Surprisingly, synaptic development is not affected by mutation of neuroligin, neurexin's most well characterized binding partner. We therefore propose that neurexin expression in cholinergic neurons organizes synaptic connectivity with GABAergic neurons through a neuroligin-independent trans-synaptic signaling pathway. Our findings provide evidence that cholinergic neurons utilize distinct molecular signals to govern the establishment of synaptic connectivity with GABAergic motor neurons and body wall muscles, offering a novel view into molecular mechanisms responsible for generating divergent patterns of synaptic connectivity.

Disclosures: M.L. Lemons: None. **A. Philbrook:** None. **S. Ramachandran:** None. **D. Oliver:** None. **C. Lambert:** None. **M.M. Francis:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.21/B46

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Title: Ultrasonic vocalization and sociability deficits in mice lacking SRPX2

Authors: *B. SOTEROS[1], G.-M. SIA[2]
[1]Pharmacol. Dept, UT Hlth. San Antonio, San Antonio, TX; [2]Pharmacol., Univ. of Texas Hlth. Sci. Ctr. at San Antonio, San Antonio, TX

Abstract: Synapse formation and maintenance are critical for the development of proper neural circuitry. SRPX2, the sushi-repeat containing domain protein x-linked 2, is a human language associated protein which regulates synapse formation and vocalizations in mice. We report here that mice lacking SRPX2 display deficits in ultrasonic vocalization and sociability. SRPX2-KO mice also show a lack of preference for social novelty in the three-chamber sociability task. SRPX2 is broadly expressed in the brain and is enriched in the cortex. Deletion of SRPX2 results in a reduction in VGLUT2 staining in cortical layer IV. Golgi staining revealed reduced spine density in layer V cortical pyramidal neurons, specifically on the apical dendrite which extends through layer IV. Abnormalities in cortical spine density are observed as early as P6, which parallels the onset of deficits in ultrasonic vocalization. These studies suggest that SRPX2 is important for the development of ultrasonic vocalization and cortical synapse formation in mice. Future studies are necessary to determine whether SRPX2 regulates synapse formation in vocalization-related circuitry in mice.

Disclosures: B. Soteros: None. **G. Sia:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.22/B47

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: CIHR

Title: The Ig transmembrane protein Borderless is required for synaptic development and function in the *Drosophila* visual system

Authors: *H. S. SHAW, S. A. CAMERON, W. CHANG, Y. RAO
Neurol. and Neurosurg., McGill Univ. Hlth. Ctr., Montreal, QC, Canada

Abstract: The fruit fly visual system is an excellent model to study the general mechanisms by which cell adhesion molecules regulate synaptic development and function. Recently, our lab has identified a conserved homophilic cell adhesion molecule Borderless (Bdl). Down-regulation of Bdl is required for photoreceptor axonal tiling and targeting. The *in vivo* function of Bdl in photoreceptor axons, however, remains unknown. Interestingly, IgSF9B, the mammalian homolog of Bdl, has recently been shown to contribute to inhibitory synapse formation *in vitro*. To determine the potential role of Bdl in photoreceptor axons, we developed a phototactic behavioral assay and showed that loss of *bdl* impaired the fruit fly's ability to detect blue and green light. Since detection of blue and green light in the fly visual system is mediated by R8 photoreceptors, these results suggest a role for Bdl in the control of R8 connectivity and/or function. To further determine the exact role of Bdl in R8 axons, we examined the effects of *bdl*

mutations on synaptic structures. We found that in the absence of Bdl, synaptic vesicles were mislocalized to the non-synaptic section of the R8 axon. We then performed cell-type-specific rescue and genetic mosaic analysis, which showed that Bdl is required both pre- and post-synaptically for proper localization of synaptic vesicles. These results support a model in which Bdl acts in a homophilic manner to facilitate the interactions between R8 axon and its target neurons for the assembly and function of presynaptic complexes.

Disclosures: H.S. Shaw: None. **S.A. Cameron:** None. **W. Chang:** None. **Y. Rao:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.23/B48

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Title: Proteolytic regulation of GABAergic synapse formation by neuroligin-2

Authors: *T. TOMITA, Y. NAO, R. NAGATOMO, K. YAMASHITA, M. KIMURA
The Univ. of Tokyo, Tokyo, Japan

Abstract: Neuroligin (NL) is one of the most characterized postsynaptic cell adhesion molecules. NL binds to presynaptic partner neurexin, and plays a role for synapse formation and maturation during development. Neuroligin-2 (NL2) locates at inhibitory postsynapse, and associates with molecules involved in the inhibitory postsynaptic assembly, such as gephyrin and collybistin. NL2 induces the GABAergic synapse formation and maturation in heterologous culture system. However, relationship between its protein metabolism and synaptogenic activity is not well-understood. We have previously described the proteolytic regulation of NL1-mediated excitatory synapse formation by sequential cleavages by ADAM10 and gamma-secretase (Suzuki et al., Neuron 2012). Here, we analyzed the proteolytic cleavage and function of NL2. In rat primary neuronal culture, soluble N-terminal fragment of NL2 (sNL2) and a membrane-associated C-terminal fragment in cultured medium and cell lysates, respectively, were detected. Production of sNL2 was diminished upon treatment of metalloproteinase inhibitors, TAPI2 and GM6001, but not by ADAM protease inhibitor INCB3619. Then we screened the effect of membrane associated matrix metalloproteinases. We found that NL2 shedding was augmented and decreased by co-expression or RNAi knockdown of MT3-MMP/MMP16, respectively, suggesting that MMP16 is involved in the shedding of NL2 in primary neurons. Systematic mutagenesis analyses revealed that the stalk region of NL2 was shed, and cell surface level as well as synaptogenic activity of NL2 was correlated with the shedding efficiency. Finally, we found that GABAergic activation modulates the shedding of NL2 in primary neurons. From these results, we speculate that NL2 is proteolytically processed in the brain, and this cleavage regulates the NL2 protein level and its synaptogenic function.

Disclosures: **T. Tomita:** None. **Y. Nao:** None. **R. Nagatomo:** None. **K. Yamashita:** None. **M. Kimura:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.24/B49

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: ERC310021

Title: Highly selective cell-type programs regulate synaptic target specificity

Authors: R. DEOGRACIAS, E. FAVUZZI, A. MARQUES-SMITH, D. EXPOSITO-ALONSO, P. MAESO, *B. RICO
King's Col. London, London, United Kingdom

Abstract: Neuronal circuitries underlying the function of the mammalian cerebral cortex collectively constitute one of the most complex biological systems. As such, unraveling the mechanisms that control their development represents one of the most challenging questions in science. During development, the specificity of neuronal wiring is achieved through the coordination of multiple cues, which first guide axons to the right target area, then to the proper cellular partner and, finally, to the precise subcellular compartment onto which synapses will be formed. Subcellular segregation of synapses occurs for all types of inputs, but it reaches its highest diversity for inhibitory GABAergic terminals. Much is known about the general machinery controlling axon guidance in the developing brain; in contrast, the mechanisms of synapse segregation remain largely unknown. To understand the molecular codes that control GABAergic synaptic target specificity, we combined cell sorting approaches and high-throughput RNA-sequencing techniques to obtain the transcriptome profile of different interneurons populations during synapse formation. Our unbiased screening has unveiled different candidate genes that could be putatively involved in synapse formation. Using a structural and functional analysis in vivo, we revealed selective cell-type unique programs that regulate GABAergic synapse formation onto pyramidal neurons.

Disclosures: **R. Deogracias:** None. **E. Favuzzi:** None. **A. Marques-Smith:** None. **D. Exposito-Alonso:** None. **P. Maeso:** None. **B. Rico:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.25/B50

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: NIH Grant R01 MH097949

NIH Grant K99 MH110665

Title: Kit, an RTK linked to neurodevelopmental disabilities, regulates the specification of connectivity in the developing murine cerebellum

Authors: *M. R. WILLIAMS[1], C. P. RIZZUTO[2], J. LEE[2], B. W. LUIKART[3]
[1]Physiol. & Neurobio., Dartmouth Med. Sch., Lebanon, NH; [2]Dartmouth Col., Lebanon, NH; [3]Mol. and Systems Biol., Dartmouth Col. Geisel Sch. of Med., Lebanon, NH

Abstract: Autism spectrum disorder (ASD) has a large genetic component. Our objective is to understand the role of ASD candidate genes in brain development and ultimately identify therapeutic targets. Here, we study the receptor tyrosine kinase, Kit (KitR). KitR mutations are rarer than would be expected in the population, suggesting a deleterious effect of its loss of function. Indeed, in case studies with known or suspected KitR mutations, children present with profound syndromic ASD and/or intellectual disability. KitR is enriched in brain areas germane to the symptoms of ASD, including hippocampus (learning and memory), striatum (repetitive behaviors), and cerebellum (sensorimotor learning and function). It has been known that KitR and its ligand KitL are expressed in opposing synaptically coupled cells within these circuits. This pattern suggests that KitL/KitR may influence synaptogenesis in these structures. If so, loss of KitR may disrupt the development and dependent behaviors of these circuits, contributing to the emergence of ASD. Here, we begin testing this model by determining if KitL attracts innervation by KitR expressing cells during development. In human and mouse cerebellum, KitL is abundantly expressed by Purkinje cells (PCs) and KitR is expressed by Molecular Layer Interneurons (MLIs). In development, MLIs migrate to and synapse upon Purkinje cells. We have used viral and transgenic strategies to manipulate KitL and KitR in the developing mouse cerebellum, and we have used electrophysiology and immunohistochemistry to study the impacts on MLI migration, morphology, and synaptogenesis. We find that the ectopic expression of KitL is sufficient to attract the migration of and functional innervation by MLIs, and that the MLI expression of Kit is necessary for the normal innervation of PCs. We also provide evidence that the expressivity of KitL and KitR throughout the brain support the model that Kit signaling modulates the patterning of synaptic connectivity in the developing brain.

Disclosures: **M.R. Williams:** None. **C.P. Rizzuto:** None. **J. Lee:** None. **B.W. Luikart:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.26/B51

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: CIHR

NSERC

FRQS

Title: Shunting $GABA_A$ transmission restrains glutamtergic synapse formation in the developing hippocampus

Authors: *C. K. SALMON[1], H. PRIBIAG[2], G. QUESSEVEUR[3], M. A. WOODIN[4], K. K. MURAI[1]
[1]Ctr. For Res. In Neurosci., Montreal, QC, Canada; [2]UCSD, LA Jolla, CA; [3]Ctr. for Res. in Neurosci., Montreal, QC, Canada; [4]Univ. of Toronto, Toronto, ON, Canada

Abstract: GABA is the main inhibitory neurotransmitter in mature neurons, but depolarizes immature neurons during development. GABA's early depolarizing action controls numerous aspects of circuit formation, in particular glutamatergic synapse formation and refinement. Interestingly, during the peak of gluatmatergic synapse formation in the hippocampus, $GABA_A$ transmission changes from depolarizing, to shunting, to hyperpolarizing. Thus, the question of timing is central when probing the role of GABAergic transmission in circuit development. We therefore asked the question, how does $GABA_A$ transmission regulate glutamatergic synapse formation as it shifts from depolarizing to fully inhibitory. To do so we turned first to the organotypic hippocampal slice. As the literature suggests, blocking hyperpolarizing GABA transmission for 48 hours in these cultures caused loss of dendritic spines due to over-excitation. In contrast, blocking GABA transmission for 48 hours just prior to the switch, when GABA is shunting, caused a marked increase in dendritic spines, synaptic proteins and mEPSC frequency. These changes lasted for up to a week following washout of the GABA blockade and were associated with elevated BDNF and cFos mRNA levels. Furthermore, slices from mice heterozygous for BDNF fail to increase expression of synaptic markers following GABA blockade over the same period of time. Our work emphasizes the necessity for intact $GABA_A$ transmission during nervous system development and uncovers a narrow developmental time window in which GABA restrains synapse formation. Moving forward, to investigate whether this phenomenon occurs *in vivo*, we established methods for TetOn-based doxycycline-mediated gene expression using *in utero* electroporation, and also developed a panel of doxycycline-

inducible GABA-disrupting plasmids. Using these tools we will assess the role of $GABA_A$ transmission in developmental synapse formation with temporal and spatial precision *in vivo*.

Disclosures: C.K. Salmon: None. **H. Pribiag:** None. **G. Quesseveur:** None. **M.A. Woodin:** None. **K.K. Murai:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.27/B52

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: JSPS 17H05767

Title: Decreased cohesin in the brain leads to defective synapse development and anxiety-related behavior

Authors: *Y. FUJITA, T. YAMASHITA
Dept Mol Neurosci, Grad Sch. Med, Osaka Univ., Suita, Osaka, Japan

Abstract: The cohesin complex, composed of four subunits, including Smc3, plays a role in sister-chromatid cohesion. Although expression of cohesin in dividing cells is well established, cohesin is also expressed in post-mitotic cells. Recent studies demonstrate that cohesin is involved in forming chromatin loops at particular loci and regulates gene expression in vitro. However, if cohesin plays a role in terminally differentiated cells in vivo is unknown. Therefore, we generated conditional Smc3-knockout mice and examined their brain phenotype. We observed greater dendritic complexity and larger numbers of immature synapses in the cerebral cortex of Smc3 heterozygous deficient mice than in wild-type mice. Neuron-specific Smc3-knockout mice showed the same phenotype, indicating that Cohesin function in neurons are required for proper neuronal network formation. Smc3 heterozygous deficient mice also exhibited more anxiety-related behavior, which is a symptom of Cornelia de Lange syndrome. Further, a gene-ontology analysis following RNA sequencing suggested the enrichment of immune processes, particularly the response to interferons, in the Smc3 heterozygous deficient mice. Indeed, fewer synapses formed in their cortical neurons, and this phenotype was rescued by STAT1 knockdown. Thus, low levels of cohesin expression in the developing brain leads to changes in gene expression that in turn lead to a specific and abnormal neuronal and behavioral phenotype.

Disclosures: Y. Fujita: None. **T. Yamashita:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.28/B53

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: JSPS Grants-in-AID

HFSP Program Grant

Title: Development of a novel designer synapse connector and control of synapse formation and behavior *In vivo*

Authors: *K. SUZUKI[1], W. KAKEGAWA[1], E. MIURA[1], J. ELEGHEERT[2], A. CLAYTON[2], R. KAUSHIK[3], A. DITYATEV[3], R. A. ARICESCU[2,4], M. YUZAKI[1]
[1]Dept. of Neurophysiol., Keio Univ. Sch. of Med., Tokyo, Japan; [2]Univ. of Oxford, Oxford, United Kingdom; [3]German Ctr. for Neurodegenerative Dis., Magdeburg, Germany; [4]MRC Lab. of Mol. Biol., Cambridge, United Kingdom

Abstract: Synapse formation and its maintenance accomplished by synapse organizers are crucial for the proper development of the brain and their disruption would lead to neuropsychiatric diseases so called “synapse diseases”. Extracellular scaffolding proteins (ESPs), such as cerebellin 1 (CBLN1) and neuronal pentraxin 1 (NPTX1), are the recently identified new class of synaptic organizers. It is noteworthy that the application of recombinant ESPs has been shown to rapidly regulate synapse formation and plasticity *in vitro* and *in vivo*, indicating that ESPs could serve as potential tools to arbitrarily control synapse formation and its function. Here we developed a novel synapse connector protein, Cerebellin-Pentraxin chimeric protein (CPTX) which is composed of the stalk domain of CBLN1 and the pentraxin domain of NPTX1. CPTX is designed to make selective and simultaneous connection between presynaptic neurexins with splice site 4 and postsynaptic AMPA receptors via respective domains. As expected, CPTX specifically bound to HEK cells expressing neurexins with splice site 4 and N-terminal domain of AMPA receptor subunits (GluA1-4). Treatment of CPTX *in vitro* induced the synapse formation between either CBLN1-null cerebellar granule neurons or wild-type hippocampal neurons and HEK cells expressing the N-terminal domain of AMPA receptor. CPTX accumulated between pre- and postsynaptic elements *in vitro* and *in vivo*. To examine the functional effect of CPTX *in vivo*, we tried to rescue motor discoordination in two different model mice, CBLN1-null and GluD2-null mice, both of which show ataxia because of the few synapse formation in cerebellum. Remarkably, CPTX injection into cerebellar vermis of the either mouse rescued the motor discoordination. Furthermore, electron microscopic and electrophysiological analysis revealed that CPTX injection restored functional synapse formation in both mice. These results suggest that CPTX can connect excitatory synapses and modulate the

motor performance. This research would lead to potential new avenues for the treatment of "synapse diseases".

Disclosures: **K. Suzuki:** None. **W. Kakegawa:** None. **E. Miura:** None. **J. Elegheert:** None. **A. Clayton:** None. **R. Kaushik:** None. **A. Dityatev:** None. **R.A. Aricescu:** None. **M. Yuzaki:** None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.29/B54

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: NIH NS050356

SFARI-177037

Title: Neurexin, neuroligin and wishful thinking in synaptic cytoarchitecture and growth at NMJ

Authors: *S. BANERJEE
Physiol., UT Hlth. Sci. Ctr. At San Antonio, San Antonio, TX

Abstract: Trans-synaptic interactions involving Neurexins and Neuroligins are thought to promote adhesive interactions for precise alignment of the pre- and postsynaptic compartments and organize synaptic macromolecular complexes across species. In *Drosophila*, while Neurexin (Dnrx) and Neuroligins (Dnlg) are emerging as central organizing molecules at synapses, very little is known of the spectrum of proteins that might be recruited to the Dnrx/Dnlg trans-synaptic interface for organization and growth of the synapses. Using full length and truncated forms of Dnrx and Dnlg1 together with cell biological analyses and genetic interactions, we report novel functions of Dnrx and Dnlg1 in clustering of pre- and postsynaptic proteins, coordination of synaptic growth and ultrastructural organization. We show that Dnrx and Dnlg1 extracellular and intracellular regions are required for proper synaptic growth and localization of Dnlg1 and Dnrx, respectively. *dnrx* and *dnlg1* single and double mutants display altered subcellular distribution of Discs large (Dlg), which is the homolog of mammalian post-synaptic density protein, PSD95. *dnrx* and *dnlg1* mutants also display ultrastructural defects ranging from abnormal active zones, misformed pre- and post-synaptic areas with underdeveloped subsynaptic reticulum. Interestingly, *dnrx* and *dnlg1* mutants have reduced levels of the Bone Morphogenetic Protein (BMP) receptor Wishful thinking (Wit), and Dnrx and Dnlg1 are required for proper localization and stability of Wit. In addition, the synaptic overgrowth phenotype resulting from the overexpression of Dnrx fails to manifest in *wit* mutants. Phenotypic analyses of *dnrx*/*wit* and *dnlg1*/*wit* mutants indicate that Dnrx/Dnlg1/Wit coordinate synaptic growth and architecture at

the NMJ. Our findings also demonstrate that loss of Dnrx and Dnlg1 leads to decreased levels of the BMP co-receptor, Thickveins and the downstream effector phosphorylated Mad at the Neuromuscular Junction (NMJ) synapses indicating that Dnrx/Dnlg1 regulate components of the BMP signaling pathway. Together our findings reveal that Dnrx/Dnlg are at the core of a highly orchestrated process that combines adhesive and signaling mechanisms to ensure proper synaptic organization and growth during NMJ development.

Disclosures: S. Banerjee: None.

Poster

368. Synapse Formation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 368.30/B55

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: Vlaams Instituut voor Biotechnologie (VIB)

Fonds Wetenschappelijk Onderzoek - Vlaanderen (FWO)

Belgian Science Policy Office (BELSPO)

Swiss National Science Foundation (SNSF)

Title: Compartmentalized regulation of the InR/PI3K pathway by the Prl phosphatase directs spatial specificity of CNS synaptogenesis

Authors: *O. URWYLER[1,2], A. IZADIFAR[2], S. VANDENBOGAERDE[2], K. VINTS[3], A. KREMER[3], D. SCHMUCKER[2]
[1]Inst. of Mol. Life Sci., Univ. of Zürich, Zürich, Switzerland; [2]VIB & KU Leuven, Leuven, Belgium; [3]VIB BioImaging Core, Leuven / Gent, Belgium

Abstract: Precise subcellular and spatial control of synapse number or synapse type is of central importance for CNS circuit development. We use genetic tools and correlative light and electron microscopy (CLEM) to investigate synaptogenesis at single-neuron resolution in the *Drosophila* CNS. In an *in vivo* RNAi screen and subsequent validation with loss-of-function mutants, we uncovered a role for Phosphatase of regenerating liver (Prl-1) specifically in spatially restricted formation of synapses. Loss of Prl-1 strongly reduces the number of presynaptic endings at terminal axonal arborizations but not the number of en-passant synapses. Our results indicate that the synaptogenic function of Prl-1 involves an axon branch specific regulation of the Insulin Receptor - Pi3K pathway, as well as compartmentalized activation of Akt. We propose a model of the cellular and molecular mechanisms for Prl-1 mediated modulation of signaling pathways that control spatial specificity of synapse formation in the developing CNS.

Disclosures: **O. Urwyler:** None. **A. Izadifar:** None. **S. Vandenbogaerde:** None. **K. Vints:** None. **A. Kremer:** None. **D. Schmucker:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.01/B56

Topic: A.07. Developmental Disorders

Support: Seaver Foundation

Title: Electrophysiological correlates of simulated language acquisition in autism spectrum disorder

Authors: *E. ISENSTEIN[1], A. KEY[2], J. H. FOSS-FEIG[1]
[1]Seaver Autism Ctr., New York, NY; [2]Dept. of Hearing and Speech Sci., Kennedy Ctr. for Res. on Human Develop., Nashville, TN

Abstract: BACKGROUND: Autism spectrum disorder (ASD) is a neurodevelopmental disorder affecting 1 in 68 children. Almost 40% of affected individuals never develop speech, and lack of language skills associates with cognitive impairment and poorer functional outcomes in this population. Therefore, understanding the neural basis of language learning in ASD can provide insight into a core deficit. It also may inform who will benefit from certain language interventions and treatment options.
METHOD: EEG data were recorded continuously at 1000Hz using a 128-channel geodesic net for 28 individuals (15 with ASD; 13 typically developing (TD)) who heard prerecorded “non-word” stimuli during a passive-listening task meant to simulate language learning. A randomly selected non-word was repeated 50 times throughout the paradigm; all other non-words were presented once. Following pre-processing procedures, amplitude and latency for the positive deflection 250-550ms after stimulus onset were extracted over temporal-parietal electrodes, separately for Repeated and Single stimuli.
RESULTS: Both groups (ASD, TD) showed significant differentiation of neural response to Repeated versus Single non-word stimuli: both showed greater amplitude in the Repeated condition (main effect, $F(1,26)=5.34$, $p<.05$). However, latency differences were observed between groups. Across Conditions, latency was faster in ASD, (main effect of Group, $F(1,26)=10.85$, $p<.01$). However, whereas latency was faster to Repeated versus Single non-words in TD, latency was slower to Repeated vs. Single non-words in ASD (Group-by-Condition Interaction, $F(1,26)=5.41$, $p<.05$).
CONCLUSION: Our results demonstrate significant differences between ASD and TD in the electrophysiological response to language-like stimuli. In TD, neural response is quicker for repeated stimuli, likely reflecting a learning effect wherein repeated (learned) words are

processed more efficiently. In ASD, however, response latency was slower to repeated non-word stimuli. This finding could indicate an attention bias to novel words and, consequently, fewer cognitive resources for processing repeated words. The end result of such a bias may be slower, less efficient processing of stimuli to be "learned." Notably, both groups showed greater amplitude to Repeated non-word stimuli, demonstrating that language learning is associated with more robust neural response regardless of diagnosis. Coupled with this amplitude finding, the ASD latency finding could suggest that language learning (i.e. response to repeated non-word stimuli) occurs in ASD, but is associated with relative slowing of neural processing.

Disclosures: E. Isenstein: None. **A. Key:** None. **J.H. Foss-Feig:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.02/B57

Topic: A.07. Developmental Disorders

Support: Brain and Behavior Research Foundation NARSAD Young Investigator

NICHD U54 HD090256

NICHD P30 HD003352

Title: Microstructure and morphometry of the pons in response to balance training in individuals with autism

Authors: *I. GALLAGHER[1], A. GOMEZ[1], O. DADALKO[1], K. MCLAUGHLIN[1], O. J. SURGENT[1], B. KOEHN[1], B. TRAVERS[2,3,1]
[1]Waisman Ctr., Madison, WI; [2]Occup. Therapy Program in the Dept. of Kinesiology, Madison, WI; [3]Univ. of Wisconsin-Madison, Madison, WI

Abstract: Background: Autism spectrum disorder (ASD) is a neurodevelopmental disorder defined by continuous deficits in both verbal and nonverbal communication, and the presence of restricted and/or repetitive behaviors. Individuals with ASD have been shown to exhibit challenges in postural stability that have been positively associated with autism symptom severity. The pons region of the brainstem is known to subserve postural stability, but it is unclear how the pons microstructure and/or volume is associated with postural stability in ASD, and whether a balance-training intervention changes neurobiological properties of the pons. **Objective**: To examine how the pons morphometry, myelin volume fraction, and tissue microstructure relate to postural stability in ASD before and after intensive balance training. **Methods**: The study involved over 30 adolescents with ASD and typical development (TD) who participated in 6-week balance training (balance group) or sedentary computer games (control

group). Postural sway area was calculated under eyes-open, eyes-closed, and biofeedback conditions. High-resolution structural MPnRAGE scans measured morphometric properties; brainstem-optimized DWI and McDESPOT images assessed tissue microstructure and myelin volume fraction, respectively.
Results: Within the ASD group, morphometry of the pons was insignificantly related to the postural stability measures, but myelin volume fraction of the pons showed significant association with mean sway area. Tissue microstructural properties of the pons associated with postural stability measures pre- and post-balance training differently in the ASD group compared to TD individuals.
Conclusions: The results indicate that in ASD higher myelination but not mean volume is related to more postural sway (i.e., poorer balance). Considering that higher myelination is positively associated with balance in TD individuals, our results suggest that there may be a compensatory developmental mechanism in ASD. Future research will focus on investigating what subregions within the pons (i.e., gray matter nuclei or white matter substructures) relate to postural stability in individuals with ASD and how tissue microstructural properties may change with balance training in ASD and typical development.

Disclosures: I. Gallagher: None. **A. Gomez:** None. **O. Dadalko:** None. **K. Mclaughlin:** None. **O.J. Surgent:** None. **B. Koehn:** None. **B. Travers:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.03/B58

Topic: A.07. Developmental Disorders

Title: Comparative study of supression of neuroinflammation by resveratrol and curcumin in the experimental paradigm of autism spectrum disorders

Authors: *R. BHANDARI[1], A. KUHAD[2]
[1]Panjab Univ., Chandigarh, India; [2]Panjab Univ., CHANDIGARH, India

Abstract: Objective: Neuronal dysfunction caused as a result of neuroinflammation triggered by the stimulation of matrix metalloproteinases and the subsequent release of pro-inflammatory cytokines is one of the probable mechanisms involved in the pathogenesis of autism spectrum disorders (ASD). The aim of the present study was to compare the ameliorative potential of resveratrol and curcumin on neuroinflammation in the experimental paradigm of neuroinflammatory model of ASD in rats
Method: 1M Propanoic acid (PPA)(4µl) was infused over 10 minutes into the anterior portion of the lateral ventricle to induce ASD like symptoms in rats. Resveratrol (5, 10 and 15 mg/kg) as well as curcumin (50, 100 and 200 mg/kg) was administered starting from the 2nd day of the

surgery and continued upto 28th day. Rats were tested for various behavioural paradigms such as social interaction, stereotypy, locomotor activity, anxiety and novelty, depression, spatial learning and memory, repetitive and pervasive behaviour between the 7th day and 28th day. In addition, biochemical tests for oxidative stress, mitochondrial complexes, TNF-α and MMP-9 were also assessed.
Results: Intracerebroventricular injection of propanoic acid produced neurological, sensory, behavioural, biochemical and molecular deficits which were assessed as endophenotypes of autism spectrum disorders. Treatment with resveratrol as well as curcumin for four weeks restored, significantly and dose dependently, all these endophenotypes in PPA induced ASD in rats. It was found that curcumin is more potent antioxidant even at low doses as indicated from its ability to reduce nitrite levels, Complex 4 activity as well as GSH levels. Also its effectiveness in restoring sensorimotor dysfunction, locomotor activity as well as memory and pervasiveness as compared to resveratrol could give an indication of this.
Conclusion: The major finding of the study is that though both resveratrol as well as curcumin could restore the core and associated symptoms of autistic phenotype by suppressing oxidative-nitrosative stress, mitochondrial dysfunction, TNF-α and MMP-9 expression in PPA induced ASD in rats. However, on comparison curcumin appears to be more potent antioxident but it needs further investigation.
Keywords: Autism spectrum disorders (ASD), resveratrol, neurobehavioural, oxido-nitrosative stress, TNF-α, MMP-9, curcumin

Disclosures: **R. Bhandari:** None. **A. Kuhad:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.04/B59

Topic: A.07. Developmental Disorders

Support: Hartwell Foundation Individual Biomedical Award

Brain and Behavior Research Foundation NARSAD Young Investigator Award

NICHD U54 HD090256

NICHD P30 HD003352

Title: Sensory challenges in autism are associated with microstructural atypicalities in the brainstem

Authors: *B. TRAVERS[1,2], O. DADALKO[3], K. AUSDERAU[3,2], K. MCLAUGHLIN[3]
[2]Occup. Therapy Program in the Dept. of Kinesiology, [3]Waisman Ctr., [1]Univ. of Wisconsin-Madison, Madison, WI

Abstract: Background: Autism spectrum disorder (ASD) is a neurodevelopmental disorder effecting 1.5% of the US population. An estimated 40-90% of individuals with ASD experience debilitating sensory symptoms, including hypo- and hyper-responsiveness as well as sensory seeking and enhanced perception. Neurobiological causes of such behavioral impairments are not well understood, encumbering the search for effective treatments. Lesion studies and research in animal models suggest that sensory challenges may stem from the aberrant neurophysiology in the brainstem. Examining how brainstem substructures are associated with sensory symptoms in ASD will provide neurobiological basis for these symptoms, leading to the development of effective treatments.Objective: To examine microstructure in select white matter tracts and gray matter nuclei of the brainstem in children with ASD and how it relates to their sensory symptoms. Methods: More than 40 children with ASD and without ASD (6-10 years of age) participated in this study. High-resolution structural MPnRAGE scans were used in combination with the brainstem-optimized DWI images to achieve highly precise segmentation of the brainstem substructures. DTI indices (fractional anisotropy [FA], axial, mean, and radial diffusivity) were measured to assess tissue microstructure in select ROIs of the brainstem. Highly precise segmentation was achieved using MRTrix software. Sensory features were measured using the Sensory Experience Questionnaire, 3.0, a caregiver-report tool. Results: Within the ASD group, hypo-responsivity alone was highly associated with the microstructure of the brainstem pyramidal tracts (as measured by FA). Intriguingly, no sensory features in ASD were related to the microstructure of the medial lemniscus, which is commonly considered a sensory tract.
Conclusions: To our knowledge, this study is the first to provide a quantitative characterization of brainstem white matter microstructure in relation to the sensory challenges commonly reported in ASD. Further analyses will examine sensory features in ASD as a function of additional brainstem nuclei.

Disclosures: B. Travers: None. **O. Dadalko:** None. **K. Ausderau:** None. **K. McLaughlin:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.05/B60

Topic: A.07. Developmental Disorders

Support: R01 MH101183 to SDB

Title: Generation of microglial developmental index: Bridging the gap between mice and humans

Authors: *R. HANAMSAGAR[1], M. ALTER[2], C. L. BLOCK[3], H. SULLIVAN[4], J. L. BOLTON[5], S. D. BILBO[6]
[1]Lurie Ctr. for Autism, Massachusetts Gen. Hosp., Charlestown, MA; [2]Univ. of Pennsylvania, Philadelphia, PA; [3]Pscyhology and Neurosci., [4]Duke Univ., Durham, NC; [5]Dept. of Anat. & Neurobio., Univ. of California-Irvine, Irvine, CA; [6]Pediatrics, Harvard Med. School/MGH, Charlestown, MA

Abstract: Purpose: Considering the critical role of microglia in integration of neurons into brain, proper circuitry and synaptic pruning, there is a pressing need to understand 1) mechanisms underlying normal and abnormal microglia development, 2) their interactions with specific neuronal subsets, and 3) the utility of microglial analyses in preclinical models for human tissue. The goal was this study was (i) to analyze microglial development over time in purified mouse tissue using a data simplification strategy, (ii) to assess potential sex differences in the microglial development, (iii) to assess sex differences in the microglial response to immune challenge, and (iv) to adopt and validate via multiple measures the same data simplification strategy for analyzing heterogeneous human brain tissue.
Materials & Methods: We isolated microglia from male and female mice hippocampi from different developmental ages from embryonic day 18 (E18) to postnatal day 60 (P60). Following whole transcriptome analysis in purified microglia, we developed a microglia-specific developmental index (MDI) based on global gene expression patterns. Specifically, genes were identified based on whether they were significantly up- or down-regulated during development and gene expression was scaled to equally weigh all genes. MDI was calculated by taking the ratio of the averaged scaled expression of all developmentally up-regulated genes and that of all the down-regulated genes for each sample. To measure microglial development in human brain tissue samples, genes common between purified microglial transcriptome and the human brain transcriptome datasets were identified. Developmentally regulated genes were identified and a sub-index for each human sample as calculated as above.
Results: MDI tracked with chronological age as expected in purified microglia. As microglia developed, the expression of immune-related molecules increased. At P60, female microglia were more developed than male microglia, but male microglia were more immune reactive than female microglia. Using multiple validation tests, we showed that MDI derived from mice can be applied to human brain transcriptome datasets. Upon applying MDI to human transcriptome datasets of autism, we found that in the diseased brain, microglial development is significantly accelerated compared to controls.
Conclusion: In conclusion, we show that microglial development is closely related to immune activity and can be used to determine the immunoreactive state in healthy and diseased human brain.

Disclosures: R. Hanamsagar: None. **M. Alter:** None. **C.L. Block:** None. **H. Sullivan:** None. **J.L. Bolton:** None. **S.D. Bilbo:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.06/B61

Topic: A.07. Developmental Disorders

Title: Age-related cortical thickness differences in adults with autism spectrum disorder

Authors: *C. RIECKEN[1], *C. RIECKEN[1], B. B. BRADEN[2]
[1]Speech and Hearing Sci., Col. of Hlth. Solutions, Tempe, AZ; [2]Dept. of Speech and Hearing Sci., Arizona State Univ. - Tempe Campus, Tempe, AZ

Abstract: Age-Related Cortical Thickness Differences in Adults with Autism Spectrum Disorder
Background: Over the course of the last 30 years, autism spectrum disorder (ASD) diagnoses have grown exponentially, thus identifying a large group of aging individuals with ASD. Currently, little is known about how aging will affect these individuals on a neuroanatomical level, compared to aging in a typically developing (TD) population. Brain aging in ASD is of concern due to the anatomical overlap of ASD-related pathology and age-related cortical thinning. Both phenomenon follow an anterior-posterior severity gradient, resulting in frontal lobe vulnerability and relative sparing of the occipital lobe.
Methods: To investigate whether adults with ASD experience exacerbated brain aging, compared to TD adults, two studies were performed using available data from the Autism Brain Imaging Data Exchange (ABIDE). The first study compared differences in cortical thickness via FreeSurfer between ASD (n=16) and TD (n=14) participants in two age groups: young adults (YA), between 18 and 25 years of age (n = 30) and middle-aged (MA), from 39 to 58 years of age (n = 30). There were no significant differences in IQ between ASD and TD participants within YA and MA groups. The second study compared correlations between cortical thickness and age in adult ASD (n = 146) and TD groups (n = 160), controlling for IQ, to investigate the interaction of cortical thickness and age between the two groups.
Results: Study 1 found significant interactions between diagnosis group (ASD and TD) and age group (YA and MA) for the frontal and parietal lobes, and the interaction approached significance for the temporal lobe. As predicted, no interaction was observed for the occipital lobe. Study 2 found significant differences between diagnosis groups in the relationship between age and cortical thickness for areas of the left anterior insula, middle, inferior, and fusiform temporal gyri, and superior parietal cortex. Both studies demonstrated greater age-related cortical thinning in adults with ASD, compared to TD.
Conclusion: As predicted, adults with ASD demonstrated exacerbated age-related brain differences, as measured by cortical thickness, compared to TD. These difference largely followed the anterior-to-posterior gradient, with relative sparing of the occipital lobe. Findings

demonstrate significant age-related anatomical differences between ASD and TD individuals into middle-age years. Future work is warranted to investigate whether differences in brain age trajectories will translate to unique behavioral needs in older adults with ASD.

Disclosures: C. Riecken: None. **B.B. Braden:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.07/B62

Topic: A.07. Developmental Disorders

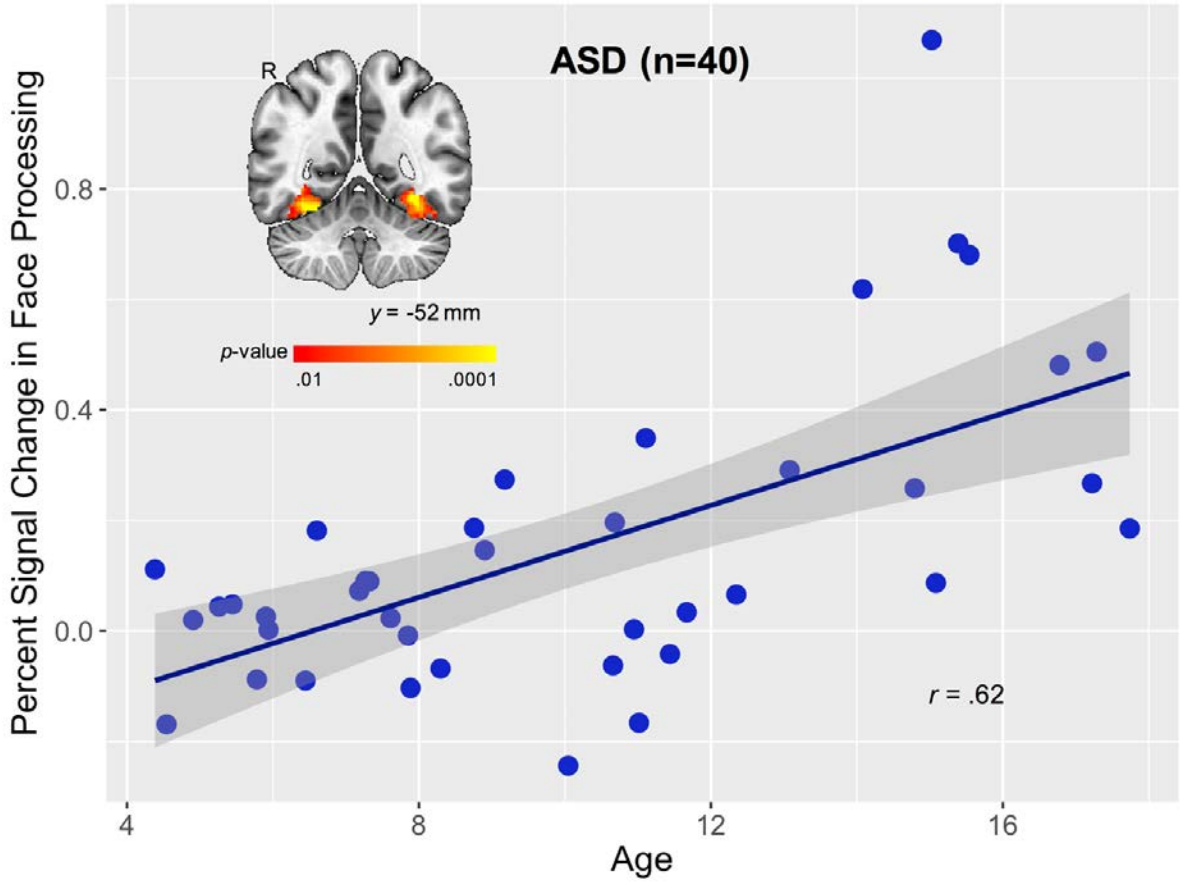
Title: Age-related developmental changes in the neural response to faces from childhood through adolescence in autism spectrum disorder

Authors: *D. YANG[1,2,3], A. WESTPHAL[3], K. A. PELPHREY[1,2]
[1]Autism & Neurodevelopmental Disorders Inst., The George Washington Univ., Ashburn, VA; [2]Children's Natl. Hlth. Syst., Washington, DC; [3]Yale Univ., New Haven, CT

Abstract: Objective: In typically-developing individuals, recent neuroscientific research demonstrates age-related growth in the face selectivity of the fusiform face area (FFA; Peelen et al., 2009, Dev Sci, 12, F16-25; Gomez et al., 2017, Science, 355(6320), 68-71). However, it remains unclear whether individuals with Autism Spectrum Disorder (ASD) exhibit similar developmental trajectories. Here, we examined age-related changes in the neural response to faces in children and adolescents with ASD.
Methods: Participants included 40 youth (10 females) diagnosed with ASD (age *M*=10.18, *SD*=4.03; IQ *M*=95.47, *SD*=23.32), characterized using gold-standard diagnostic tests. During fMRI at 3T, participants viewed alternating 5 blocks of fearful faces (FACE; 12 sec per block) and 5 blocks of houses (HOUSE; 12 sec per block). The fixation (FIX) at the beginning (10 sec) and end (20 sec) was treated as an implicit baseline. The group-level fMRI analysis utilized mixed-effects modeling and was thresholded at $Z>2.33$ (voxel) and $p<.05$ (cluster), with IQ and sex as covariates of no interest.
Results: Across the whole brain, on average, neural response to FACE>FIX was observed in the fusiform gyrus, while the neural response to HOUSE>FIX was observed in the parahippocampal gyrus. Moreover, the whole-brain analysis showed that age was linearly and positively correlated with the level of activation to FACE>FIX specifically in the fusiform gyrus. The scatterplot between age and activation within the region is illustrated in **Figure 1**. In contrast, there were no age-related neural correlates with the contrast of HOUSE>FIX across the whole brain.
Conclusions: The results revealed that in ASD, there was an age-related increase in the neural response to FACE>FIX from childhood through adolescence (4-17 years of age) in the fusiform gyrus. The results are specific to this contrast and this brain region, as there were no age-related

correlates in the contrast of HOUSE>FIX, and the age correlates were seen particularly in the fusiform gyrus, but not elsewhere in the brain.

Figure 1:



Disclosures: D. Yang: None. **A. Westphal:** None. **K.A. Pelphrey:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.08/B63

Topic: A.07. Developmental Disorders

Title: Neurofunctional properties of cognitive reappraisal for circumscribed interests in autism

Authors: *L. D. ANTEZANA, M. C. COFFMAN, J. A. RICHEY
Dept. of Psychology, Virginia Tech., Blacksburg, VA

Abstract: Background: Autism spectrum disorder (ASD) is characterized by restricted and repetitive behaviors (RRB) and difficulties in social communication. Circumscribed interests (CI) are one of the key features of RRB. While it is known that individuals with ASD demonstrate increased affective responses toward CI images (e.g., hyperarousal; Sasson et al., 2012), it is unknown whether approach-related motivation is a consequence of broader deficits in executive function, particularly emotion regulation (ER). Prior neuroimaging work from our group has highlighted that social deficits in ASD may be related to impaired ER mechanisms in ASD (Richey et al., 2015), however this principle is less understood in the context of CI.
Objective: To apply an ER framework and identify specific neurofunctional impairments in ASD that may underlie emotional responses to CI images using fMRI during ER instructions in adults with ASD versus matched controls (CON).
Hypotheses: We hypothesize that the ASD group will have difficulty modulating affect toward CI, as demonstrated by decreased dorsolateral prefrontal cortex (dlPFC) activation during ER instructions, compared to CON.
Methods: A total of 30 adults (ASD=15; CON=15) participated in this study. Groups were matched on age (ASD M=26.1; CON M=27.4) and IQ (ASD M=113.3; CON M=116.3). fMRI data were collected during an ER task on a 3T General Electric Signa Excite HD scanner. For stimuli, each individual with ASD was asked to bring ten photographs of their CI. After standardized training in cognitive ER techniques, participants viewed each image for 4 seconds (free viewing period) and were then asked to "Think Positive," or "Think Negative" about the images. To control for intra-individual variation (between ASD and CON) in the subjective level of interest in a CI image, we covaried for self-reported arousal and affective valence in our fMRI models.
Results: Whole-brain analyses revealed group differences between ASD and CON groups for think-negative vs. baseline (pre-instruction) and think-positive vs. baseline conditions ($p<0.05$). Contrary to predictions, the control group showed significantly increased activation in the precuneus and posterior cingulate cortex (PCC), compared to the ASD group for both conditions.
Conclusions: Individuals with ASD showed decreased activation in PCC, a region thought to be related to self-referential processing (Lynch et al., 2013) during ER instructions for CI. These results held after covariate adjustments for arousal and valence. These findings may indicate that modulation of self-referential processing, rather than emotion, may be linked to approach-related symptoms such as CI.

Disclosures: L.D. Antezana: None. **M.C. Coffman:** None. **J.A. Richey:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.09/B64

Topic: A.07. Developmental Disorders

Support: HIAS15004 Hussman Foundation Pilot Grant

Title: Convergent pathobiology in human iPSCs from individuals with idiopathic autism uncovered by transcriptomic and neurophysiological analysis

Authors: *M. W. NESTOR[1,2], B. DEROSA[3,4], J. EL HOKAYEM[3,4], E. ARTIMOVICH[5], C. GARCIA-SERJE[3,4], A. W. PHILLIPS[5], M. L. CUCCARO[3,6], H. N. CUKIER[3,7], J. M. VANCE[3,4,6], M. A. PERICAK-VANCE[3,6,4], D. DYKXHOORN[3,4]
[1]The Hussman Inst. For Autism, Sykesville, MD; [2]The Univ. of Maryland, Sch. Of Med., Baltimore, MD; [3]Univ. of Miami Miller Sch. of Med., Miami, FL; [4]John P. Hussman Inst. for Human Genomics, Miami, FL; [5]The Hussman Inst. for Autism, Baltimore, MD; [6]John T. Macdonald Fndn. Dept. of Human Genet., Miami, FL; [7]Dept. of Neurol., Miami, FL

Abstract: Background: Autism spectrum disorders (ASDs) are a phenotypically and etiologically complex group of neurodevelopmental conditions that are broadly behaviorally defined as clinical phenotypes involving communicative, social, and behavioral impairments. There has been considerable lag in progress made towards understanding the relationship between suspected disease gene variants in ASD and how these variants result in the cell-based phenotypes to which they are associated.
Objective: The aim of this study is to identify key differences in the function of important neurobiological processes, and in the expression patterns of molecular gene networks that regulate them, both in a temporal and neural-specific manner.
Methods: We examined the transcriptional differences between cortical neurons from patients with idiopathic ASD and healthy unaffected control individuals over the course of their in vitro development. We derived iPSCs from peripheral blood mononuclear cells (PBMCs) from idiopathic ASD individuals (DeRosa et al., 2012) and unrelated male controls and differentiated them into cortical neurons over a 135 day time course.
Results: Transcriptional analyses of ASD and control neurons at culture days 35, 85, and 135 of their in vitro development showed ASD-specific molecular phenotypes mainly affecting pathways/networks involved in neuronal differentiation, the cytoskeletal matrix structure formation (i.e. axon guidance and cell migration), regionalization, patterning, DNA and RNA metabolism. Additionally, developing networks of neurons were interrogated with multi-electrode array (MEA) recordings, measurements of calcium transients using Fluo-4, fixed and stained for relevant morphological markers and subjected to cell migration assays at time-points aligning with our transcriptional analyses. Lines from ASD individuals demonstrated significantly decreased network spiking activity from MEA recordings as well as decreased numbers of calcium transients. Additionally, ASD lines showed significant differences in measures of neurite morphology, and decreased cell migration at early neuronal differentiation times.
Conclusions: The results of this study suggest that in iPSC-derived neurons derived from individuals with ASD, there may be early deficits in network activity and morphology based on a combination of cell based assays, including MEA, Fluo-4 measured calcium transients, and quantification of neurite outgrowth that converge with transcriptomic analyses. Taken together, this suggests that for our cohort of individuals with idiopathic autism, there are convergent pathophysiological processes.

Disclosures: **M.W. Nestor:** None. **B. DeRosa:** None. **J. El hokayem:** None. **E. Artimovich:** None. **C. Garcia-serje:** None. **A.W. Phillips:** None. **M.L. Cuccaro:** None. **H.N. Cukier:** None. **J.M. Vance:** None. **M.A. Pericak-vance:** None. **D. Dykxhoorn:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.10/B65

Topic: A.07. Developmental Disorders

Support: Nancy Lurie Marks Family Foundation

NJ Governor's Council for Autism Research and Treatments

Title: Reassessing the autistic phenotype using big-data from the autism brain imaging data exchange repository

Authors: *C. CABALLERO[1], S. MISTRY[2], C. WHYATT[3], E. B. TORRES[4]
[1]Rutgers University. Psychology Dept., Piscataway, NJ; [2]Mathematics, Rutgers Univ., Piscataway, NJ; [3]Rutgers Univ. - Busch Campus, Piscataway, NJ; [4]Psychology Dept, Rutgers Univ. Dept. of Psychology, Piscataway, NJ

Abstract: Large open-access repositories of imaging data sets have initiated a new avenue of enquiry in Autism Spectrum Disorders (ASD). Along with the gaining of access to imaging data from different cognitive and psychological experiments interspersed with resting state (RS) fMRI, it is now also possible to examine large cross-sectional demographic data. This provides unprecedented statistical power to draw inferences about many aspects of the autistic phenotype. Furthermore, several sites provide longitudinal data for the same subjects set, permitting as well the derivation of dynamic outcome measures to track disorder progression, longitudinal effects of medication intake, among other queries that such Big-Data enables. Despite these great benefits to advance basic research and enrich clinical knowledge in ASD, we face several challenges when combining data from different sites in the repository harnessed under disparate sampling resolution (SR). For example, in the Autism Brain Image Date Exchange (ABIDE) data sets some sites have SR above 1Hz while others have SR below 1Hz. Given that several parameters for statistical inference can be derived from imaging data, and queries performed according to demographic data, here we ask if the raw head-motion signal derived from time series acquired under different SR may be representing different stochastic processes according to the noise type their inherent variability has. Using methods to assess stochastic processes and derive noise signatures from head motion data in RS-fMRI, we find that sites across ABIDE I and II repositories with SR above 1Hz have range in the pink-noise regime while those below 1Hz have noise range in the Brownian-motion regime. These results caution against pooling data

from all sites to derive statistical inferences. Further, we provide a new standardized data type (amplitude fluctuations' micro-movements) insensitive to these nuances and perform several queries on sex differences in ASD, medication intake and age-dependent evolution of the stochastic signatures and their rates of change. We report statistically significant differences between (1) ASD and AS females in relation to age-matched TD females; (2) medication vs. no medication intake; (3) age-dependent rates of change in stochastic signatures and further report outcomes on stochastic assessment of longitudinal data. Our results are discussed within the context of dynamic biomarkers for Precision Medicine and Computational Psychiatry.

Disclosures: C. Caballero: None. **S. Mistry:** None. **C. Whyatt:** None. **E.B. Torres:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.11/C1

Topic: A.07. Developmental Disorders

Support: NICHD 2P50HD055784-08

Title: Early electrophysiological markers of neural network development in infants at risk for autism spectrum disorder (ASD)

Authors: *A. DICKINSON[1], E. LIN[1], A. SCHEFFLER[1], D. SENTURK[1], M. DAPRETTO[2], S. S. JESTE[1]
[2]Dpt Psych & Biobehav Sci., [1]UCLA, Los Angeles, CA

Abstract: Aberrant neural connectivity is present in adults with autism spectrum disorder (ASD), and is widely cited as a neurobiological mechanism underlying the condition (e.g. Wass, 2011). Connectivity between brain regions develops extensively throughout infancy, underlying the formation of increasingly sophisticated neural networks. Disruptions in neural networks may be evident during infancy, preceding the emergence of ASD symptoms. Investigating early brain development in ASD requires studying infants who are too young to receive a diagnosis but are considered at 'high risk', based on family history of ASD. Using this approach, we can study how early brain development differs in the proportion of children go on to develop ASD.
Large scale synchronous neuronal activity is mediated by the integrity of neural networks. Measurement of oscillatory brain activity using electroencephalography (EEG) can therefore be used to assay neural network development. Oscillations within the alpha range (6-12Hz) show clear developmental changes which reflect developing neural networks (Rodríguez-Martínez *et al.* 2017). Peak alpha frequency (PAF) and alpha band coherence are particularly sensitive to developmental change, both increasing with age during typical development (Miskovic *et al.* 2015; Gmehlin *et al.* 2011).

Here we study spontaneous EEG data collected from 31 high risk, and 29 low risk infants, at 3, 6, 9 and 12 months of age in the UCLA Center of Autism Research and Treatment. PAF was computed by removing the 1/f trend from the calculated power spectra, and fitting a Gaussian curve to accurately capture power modulations in the alpha range. EEG data were transformed into current source density to study phase coherence between electrode pairs. A permutation test with a false discovery rate adjustment for multiple testing, was used to identify electrode pairs which demonstrated significantly altered phase coherence in high risk infants.
Preliminary data analysis suggests that alterations in PAF and alpha phase coherence are present during the first year of life in infants at high risk for ASD. Lower PAF at 6 months of age is also associated with higher degree of ASD concern at 18 months of age. This suggests that variability in the integrity of neural networks during the first year may map onto later clinical heterogeneity. Further analysis will explore how individual variability in the timing and location of neural network disruptions may inform the clinical heterogeneity of ASD. We will discuss the potential utility of scalable electrophysiological markers of network development to facilitate precisely targeted interventions and prediction of outcomes.

Disclosures: A. Dickinson: None. **E. Lin:** None. **A. Scheffler:** None. **D. Senturk:** None. **M. Dapretto:** None. **S.S. Jeste:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.12/C2

Topic: A.07. Developmental Disorders

Support: NICHD 2P50HD055784-08

ARCS

Title: Functional connectivity predicts language ability in infants at risk for ASD

Authors: *X. A. TRAN[1], C. KURE LIU[1], A. DICKINSON[1], M. DAPRETTO[2], S. S. JESTE[3]
[2]Dpt Psych & Biobehav Sci., [1]UCLA, Los Angeles, CA; [3]68-237A, UCLA Semel Inst. of Biobehavioral Sci., Los Angeles, CA

Abstract: Evidence from genetics and neuroimaging studies suggests that autism spectrum disorder (ASD) is characterized by disruptions in neural connectivity. Infants with an older sibling with ASD (high risk) are at heightened risk for developing ASD. We examined electrophysiological (EEG) measures of neural connectivity in gamma band oscillations, which are critically involved in cognitive and language development, with a focus on regional coherence. We asked whether: 1) functional connectivity in the gamma band during an auditory

language processing task differs between infants with high risk (HR) for ASD and low risk (LR) infants, and 2) functional connectivity at 3 months can predict language ability at 18 months. Our study is based on an overarching hypothesis that neural connectivity is disrupted in infants at high risk for ASD, with the biggest differences found in those infants who have delayed or atypical development.
Participants included 31 HR infants and 30 LR infants from an ongoing longitudinal study of brain development in high risk infants. EEG was acquired at 3 months while each subject listened passively to a language processing task for 2 minutes. EEG was collected using NetStation, filtered at 1-50 Hz and cleaned using EEGlab. Independent component analysis was applied and non-neural components were removed. Laplacian filter was applied using the CSD toolbox. Functional connectivity (coherence) was calculated between electrode pairs using the EEGlab function “newcrossf”. Mean left frontal-parietal coherence in the gamma frequency band (30-50 Hz) was averaged across left frontal-parietal electrode pairs. Language ability was assessed at 18 months using the Mullen Scales of Early Learning (Mullen). We used independent samples t-test to compare coherence values between HR and LR groups, and Pearson’s correlations to examine the relationship between coherence and Mullen language scores.
HR infants had higher mean left frontal-parietal coherence than LR infants, which was driven by two electrode pairs – “Fp1-P3” and “F9-P9”. In the HR group, coherence in “Fp1-P3” electrode pair negatively correlated with Mullen verbal and expressive language t-scores. Correlations between coherence and language were insignificant in the LR group.
Compared to LR infants, HR infants had higher regional functional connectivity during auditory language processing at 3 months that may be evident of underlying atypical neural connectivity. Elevated functional connectivity in HR infants may serve as an early marker of atypical language development, as this negatively correlated with later language ability.

Disclosures: **X.A. Tran:** None. **C. Kure Liu:** None. **A. Dickinson:** None. **M. Dapretto:** None. **S.S. Jeste:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.13/C3

Topic: A.07. Developmental Disorders

Support: NIMH R01 MH100173 (McPartland)

CTSA Grant Number UL1 RR024139 (McPartland)

NIMH K23 MH086785 (McPartland)

NIMH R21 MH091309 (McPartland)

Autism Speaks Translational Postdoctoral Fellowship (Naples)

Waterloo Foundation 1167-1684 (McPartland)

Patterson Trust 13-002909 (McPartland)

Title: Hemispheric and developmental differences in the neural processing of mutual eye contact in individuals with autism spectrum disorder

Authors: *K. A. MCNAUGHTON, A. NAPLES, T. C. DAY, M. J. ROLISON, A. CHANG, T. MCALLISTER, J. MCPARTLAND
Child Study Center, Yale Univ., New Haven, CT

Abstract: Autism spectrum disorder (ASD) is defined by differences in processing of social information compared to individuals with typical development (TD). While these differences have been characterized in terms of neural response to static face stimuli, gaze-responsive stimuli offer an opportunity to examine social interactions in a more ecologically valid context. This study expanded on previous research identifying neural markers of dynamic mutual eye contact in adults with TD (Naples et al., 2017) by investigating the neural response to eye contact in children and adults with ASD and TD. Individuals with ASD (n=43, 7 female, age 10-32 years) and individuals with TD (n=48, 21 female, age 8-35 years) matched on age, IQ, and handedness participated in a gaze-contingent experiment with co-registered electroencephalography and eye-tracking. Participants viewed faces that responded to their gaze in four conditions: fixate on eyes, eyes open (eye:eye); fixate on eyes, mouth opens (eye:mouth); fixate on mouth, eyes open (mouth:eye); fixate on mouth, mouth opens (mouth:mouth). The N170, an event-related potential previously identified as a neural marker of eye contact, was analyzed. There was a main effect of condition for N170 amplitude (F(3, 267)=40.27, p<0.01) and N170 latency (F(3, 267)=19.99, p<0.01). N170 response was stronger and faster for mutual eye contact (eye:eye) than for the other three conditions (ps<0.01). There was also a hemisphere by diagnosis interaction for N170 amplitude (F(1, 89)=12.27, p<0.01), in which individuals with ASD had a stronger N170 response in the right hemisphere than in the left (t(42)=3.60, p<0.01), while the response for individuals with TD did not differ between hemispheres (p>0.05). There were developmental differences in latency of N170 response to mutual eye contact (eye:eye) and gaze-contingent mouth movement (mouth:mouth), as indicated by a trending interaction between age and condition (F(1, 88)=3.74, p=0.06). Children under age 16 had faster N170 responses to mutual eye contact compared to mouth movement (t(43)=4.47, p<0.01)) while individuals over age 16 did not differ in response to the two conditions (p>0.05). These results support the N170 as a selective marker for mutual eye contact. They also provide evidence that individuals with autism may demonstrate atypical lateralization patterns during processing of gaze-responsive social stimuli. Given the nature of autism as a developmental disorder and the developmental shifts in the N170 as a marker of mutual eye contact, future research is required to characterize the relationship between this neural marker and social function across the lifespan.

Disclosures: K.A. McNaughton: None. **A. Naples:** None. **T.C. Day:** None. **M.J. Rolison:** None. **A. Chang:** None. **T. McAllister:** None. **J. McPartland:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.14/C4

Topic: A.07. Developmental Disorders

Support: NIH 1R01 MH100028

NIH P41 EB015922

Title: Sex differences in the brain surface area of children with autism spectrum disorders

Authors: *C. CHEN, J. VAN HORN
USC, Los Angeles, CA

Abstract: *Background:* Autism spectrum disorder (ASD) is marked by social communication deficits and restricted repetitive behavior. Sex differences in ASD diagnosis in the U.S. are striking, with a male to female ratio of nearly 5:1 in recent years (Chen, Van Horn et al. 2016). Few research studies have examined sex differences in the neuroanatomy of children with ASD. Those that do have focused on sex differences in brain volume (Bloss and Courchesne 2007, Schumann, Bloss et al. 2010) and have low sample sizes of girls. Our goal is to map sex differences in the brain surface area of girls versus boys with ASD.
Methods: Five research institutions were involved in data collection: USC, UCLA, Seattle Children's Institute, Harvard, and Yale. The sample included 96 children with ASD (44 girls) and 82 typically developing (TD) children (40 girls), ages 7 to 17. Diagnosis was based on the Autism Diagnostic Interview-Revised (Lord, Rutter et al. 1994) and Autism Diagnostic Observation Schedule-II (Lord C. 2012). Freesurfer (Dale, Fischl et al. 1999) was used to determine brain surface area for 146 cortical regions extracted from the Destrieux atlas (Destrieux, Fischl et al. 2010). Multivariate ANOVA analysis was performed on all cortical brain regions, with sex and diagnosis as independent variables. Intracranial volume, pubertal status, handedness, socioeconomic status, and MRI scanner type served as covariates. Multiple comparisons were taken into consideration with a stringent $p<.01$.
Results: A main effect of sex was found on the right lateral occipito-temporal sulcus (rLOTS; $p=.009$), while a main effect of diagnosis was found on the right parahippocampal gyrus (rPG; $p=.003$). A sex by diagnosis interaction was found on the right triangular part of the inferior frontal gyrus (rTIFG; $p=.006$).
Conclusion: Previous research discovered that girls with ASD exhibit greater temporal gray and white matter volumes, relative to boys with ASD (Bloss and Courchesne 2007). Our results indicate that boys have greater rLOTS surface area than girls, while TD children possess greater rPG surface area than ASD children. Additionally, boys with ASD have greater rTIFG surface area compared to girls with ASD and TD boys, while TD girls have greater rTIFG relative to

girls with ASD and TD boys. In contrast to previous findings that girls with ASD have abnormally enlarged volumes in many regions, TD boys and boys with ASD display enlargement in several brain surface areas related to face processing and impulse control (Pageler, Menon et al. 2003, Aron, Robbins et al. 2004). Further research should be conducted to see how these brain regions contribute to sex differences in autistic behavior.

Disclosures: C. Chen: None. **J. Van Horn:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.15/C5

Topic: A.07. Developmental Disorders

Support: Conacyt Scholarship No. 574809 (OECM)

Title: Impact of the enriched environment on the multiunitary activity of the cerebellum in the autistic rat

Authors: *O. E. CRUZ MAGOS[1], G. HERRERA MEZA[2], J. MANZO[3], L. I. GARCIA[4], P. PACHECO[5], J. A. PEREZ, SR[6]
[1]CENTRO DE INVESTIGACIONES CEREBRALES, UNIVERSIDAD VERACRUZANA, Xalapa Enríquez, Mexico; [2]Unidad De Investigaciones Alimentarias (UNIDA) Ins, Xalapa, Mexico; [3]Univ. Veracruzana, Xalapa, Ver. Mexico, Mexico; [4]Ctr. de Investigaciones Cerebrales, Xalapa, Mexico; [5]Inst. de Investigaciones Biomédicas, Univ. Nacional Autonoma de México, Xalapa, Mexico; [6]cerebrales, UV, xalapa, Mexico

Abstract: Autism is a neural developmental disorder that is observed in children as a spectrum of behavioral alterations between 2 and 3 years of age. Although the number of studies in this regard increases annually, significant physiological manifestations that allow to make laboratory diagnoses are still unknown. To date, autism continues to be diagnosed only on the basis of subject's behavior. This is why it is relevant to find approaches to allow physiologicals determinations of the disorder. It is known that one of the structures invariably altered in autistic subjects is the cerebellum. Therefore, in this work we proposed that the encephalographic record of this structure could serve as a basis for spectrum´s physiological analysis. To do this, a posnatal autistic model was used in Wistar rats by the posnatal injection of a daily dose of 150 mg / kg of valproic acid to pups from day P6 to P12. Four groups, Controls (Ct) in standard (SE) and enriched environment (EE), and Autistic (At) in SE and EE were used. All of them had a multi-unit recording of the cerebellar vermis. Results showed that EE increased the cerebellar response amplitude in the Ct, but the At in SE presented a significant increase in the amplitude equivalent to that of Ct-AE, which was reduced when they were submitted to EE. Thus, we first

observe that it is possible to identify encephalographic variations characteristic of autism in the cerebellum cortex, and that these increased variations can return to a Ct level after the subject is submitted to an environmental enrichment program. The significance of this amplitude in the discharge of the cerebellar cortex requires further studies. Conacyt Scholarship No. 574809 (OECM); Cuerpo Academico de Neurociencias (UV-CA-28).

Disclosures: O.E. Cruz Magos: None. **G. Herrera Meza:** None. **J. Manzo:** None. **L.I. Garcia:** None. **P. Pacheco:** None. **J.A. Perez:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.16/C6

Topic: A.07. Developmental Disorders

Support: Intramural Program of the NIMH; ZIA MH002588-26.

Title: Sex differences in the resting-state functional connectivity of the cerebellum in autism spectrum disorders

Authors: *R. E. SMITH[1], J. AVERY[1], L. KENWORTHY[2], S. J. GOTTS[3], A. MARTIN[4]
[1]Lab. of Brain and Cognition, Natl. Inst. of Mental Hlth., Bethesda, MD; [2]Children's Natl. Hlth. Syst., Washington, DC; [3]Lab. of Brain and Cognition, NIMH/NIH, Bethesda, MD; [4]Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: Human neuroimaging studies have documented sex-specific differences in the development, structure, and function of the cerebellum (Nguon et al., 2005). This part of the brain, in conjunction with the cortex, is involved in both sensorimotor and socio-affective processes. These two domains of function are both impaired in autism spectrum disorders (ASD), and not surprisingly, the cerebellum has been highlighted in neuroimaging studies of males with ASD. Behavioral studies have also found sex differences in ASD prevalence and phenotype. Given what we know about the relationship between ASD and the cerebellum, and the sex differences associated with each, we were interested in whether there may be sex-dependent changes in the cerebellum's functional architecture in ASD. The aim of the current study was to explore this using resting-state functional magnetic resonance imaging (fMRI). We collected resting-state fMRI scans from 47 women (23 ASD, and 24 controls) and 120 men (56 ASD, and 64 controls), matched on age, head motion and IQ. Using a measure of global functional connectivity, we ran a linear mixed effects analysis across both groups to determine whether there was a sex-by-diagnosis interaction associated with resting state functional connectivity.
Two clusters in the left and right cerebellum exhibited a diagnosis-by-sex interaction in global

connectivity ($p < 0.005$ with small volume correction in the cerebellum). Compared to controls, females with ASD showed *hyper*connectivity between those cerebellar clusters and the cortex, whereas males with ASD showed the opposite pattern (*hypoco*nnectivity with the rest of the brain). Follow-up analyses of seed-based connectivity effects underlying these global connectivity differences (using a 6mm spherical region of interest centered on the peak voxel in each cerebellar cluster) revealed significant diagnosis-by-sex interactions between the cerebellum and several cortical regions including the right fusiform, right inferior frontal gyrus, right precentral gyrus, mid-cingulate, precuneus, middle temporal gyrus, and thalamus.
These results shed light on the sex-specific pathophysiology of ASD. This pattern of sex-dependent, aberrant cerebellar connectivity in ASD might explain some of the abnormalities in motor and socio-affective abilities seen in this population.

Disclosures: R.E. Smith: None. **J. Avery:** None. **L. Kenworthy:** None. **S.J. Gotts:** None. **A. Martin:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.17/C7

Topic: A.07. Developmental Disorders

Support: Pennsylvania Department of Health using Tobacco CURE Funds

Japan Agency for Medical Research and Development (AMED)

Title: Brain-wide mapping of the developmentally regulated expression of the oxytocin receptor in mice

Authors: *Z. T. NOLAN[1], S. HIDEMA[2], K. NISHIMORI[3], Y. KIM[4]
[1]Penn State Col. of Med., Hershey, PA; [2]Mol. Biol., Grad. Sch. of Agr. Sciences/Tohoku Un, Sendai-Shi, Japan; [3]Tohoku Univ., Sendai, Japan; [4]Col. of Medicine, Penn State Univ., Hershey, PA

Abstract: Oxytocin is a neuropeptide whose functions include the development and expression of social behavior. Impairment of oxytocin signaling has been implicated in many neurodevelopmental disorders such as autism. The oxytocin receptor (OxtR) is the main mediator of oxytocin ligand. Despite its significance, brain-wide spatial and temporal expression patterns of OxtR remain largely unknown. To investigate this, we have examined the expression patterns of oxytocin receptor positive neurons throughout the whole brain at developmental time points (P7, P14, P21, P28) and in adulthood (P56) using a transgenic reporter line (OxtR-Venus). This data was acquired using a scanning serial two-photon tomography and a data processing

pipeline that allows us to map fluorescently labeled cells throughout the entire brain with standardized 3D atlases. We found that overall expression peaks at P21. Moreover, OxtR neurons in different brain regions showed quantitatively different developmental regulation. For instance, we found that endopiriform nucleus expresses OxtR from P7 through P28 peaking at P21. The anteromedial nucleus of thalamus however, does not begin to express OxtR until P14 and continues to increase expression through P28. We then asked whether decreased oxytocin receptor positive neurons in adult is caused by programmed cell death or receptor downregulation without cell death. We tracked cumulative OxtR expression by using OxtR-cre:Ai14 (cre dependent expression of tdTomato) in different developmental periods. We did not observe a decreased number of cells in the OxtR-cre:Ai14, suggesting that OxtR expression decreased without cell death. We envision that our highly detailed OxtR expression map will guide future circuit based investigation to understand mechanism of oxytocin signaling in various behavioral assay. Our future work will be directed at determining if this developmental profile of the oxytocin receptor is altered in mouse models of autism spectrum disorder.

Disclosures: **Z.T. Nolan:** None. **S. Hidema:** None. **K. Nishimori:** None. **Y. Kim:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.18/C8

Topic: A.07. Developmental Disorders

Support: MEXT and AMED

Title: Cyclic adp-ribose and heat regulate oxytocin release via cd38 and trpm2 in the hypothalamus during social or psychological stress in mice

Authors: *S. AMINA[1,2], H. HIGASHIDA[3]
[2]MIND Inst., [1]Univ. of California, Davis, Sacramento, CA; [3]Dept. of Basic Res. on Social Recognition and Memory, Res. Ctr. for Child Mental Dev, Kanazawa Univ., Kanazawa, Japan

Abstract: CD38, a transmembrane protein that triggers proliferation and immune responses in lymphocytes. cADPR, catalyzed by CD38, was considered as an intracellular second messenger. But how it exerts its role in neurons was unknown. Using a neuronal cell line, NG108-15 mouse neuroblastoma x rat glioma hybrid cells, it was shown that extracellular application of cADPR, together with heat, activates cation influx downstream of OT receptor signaling in neuronal cells. Our data further suggested that a possible involvement of TRPM2 channels in OT release in the mammalian brain. As a next step, we investigated and found similar effect in intact mouse hypothalamus primary cell culture. In addition, we recently reported that OT release from the isolated hypothalamus of male mice in culture was enhanced by extracellular application of

cADPR. This OT release differed markedly between individual mice when they were group housed, due to stress. That is, when male mice received cage-switch stress and eliminated due to their social subclass, significantly higher levels of OT release were found in subordinates compared with ordinates. These results showed that cADPR/CD38 and heat/TRPM2 are co-regulators of OT secretion and suggests that CD38 and TRPM2 are potential therapeutic targets for OT release in psychiatric diseases caused by social stress.

Disclosures: S. Amina: None. **H. Higashida:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.19/C9

Topic: A.07. Developmental Disorders

Support: NCCIH grant R01AT006364

NCCIH grant R01AT008563

NCCIH grant P01AT006663

NCCIH grant R61AT009310

NCCIH grant R21AT008707

Title: Discriminating between attention deficit hyperactivity disorder and autism spectrum disorder using surface-base resting state functional connectivity

Authors: *M. JUNG, Y. TU, C. LANG, J. KONG
Dept. of Psychiatry, Massachusetts Gen. Hosp., Harvard Med. Sch., Charlestown, MA

Abstract: Introduction: Attention-deficit hyperactivity disorder (ADHD) and autism spectrum disorder (ASD) are highly prevalent neurodevelopmental disorders, and are often comorbid with one another. The underlying neurological mechanisms of this comorbidity, as well as the distinction between the two disorders remain unclear. This study aimed to construct diagnostic models for ADHD and ASD based on resting state functional brain connectivity using support vector machine (SVM) classification. **Methods:** Data from individuals with ASD (n = 86; mean age, 11.4 ± 2.1 years) and typical development (TD) (n = 125; mean age, 10.9 ± 1.6 years) were obtained from the Autism Brain Imaging Data (ABIDE). Data from individuals with ADHD (n = 83; mean age, 11.2 ± 1.9 years) were taken from the ADHD-200 data set (http://fcon_1000.projects.nitrc.org/indi/adhd200/). To investigate diagnostic features between ADHD and ASD, ROI-to-ROI (162x162 brain regions) functional connectivity analysis was

performed based on gray matter surface parcellation in each participant using FreeSurfer. The parcellation was based on the Desikan-Killiany parcellation atlas. A feature selection approach combining univariate t test and multivariate support vector machine-recursive feature elimination (SVM-RFE) was used to identify the most discriminative features between three groups: TD vs. ASD, TD vs. ADHD and ASD vs. ADHD. We classified the three groups (two-by-two) using a 10-fold cross-validation. **Result** The classification accuracies for discriminating TD from ASD, TD from ADHD and ASD from ADHD were 76.3% (sensitivity 82.4%, specificity 61.7%), 84.1% (sensitivity 88.0%, specificity 78.3%), and 79.1% (sensitivity 79.1%, specificity 80.1%), respectively. **Conclusions:** Our findings demonstrate the potential of discriminating ASD from ADHD, as well as ASD and ADHD from TD with surface-based resting state functional connectivity analysis. **Acknowledgement:** Jian Kong is supported by R01AT006364, R01AT008563, P01 AT006663, R61AT009310, and R21AT008707 (NIH / NCCIH). All authors declare no conflict of interest.

Disclosures: **M. Jung:** None. **Y. Tu:** None. **C. Lang:** None. **J. Kong:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.20/C10

Topic: A.07. Developmental Disorders

Support: 5R21MH107045

Title: Sex-related patterns of intrinsic brain function in females with autism

Authors: *D. FLORIS[1], M.-C. LAI[2], M. P. MILHAM[3], A. DI MARTINO[1]
[1]Dept. of Child and Adolescent Psychiatry, NYU Child Study Ctr., NYU, New York, NY; [2]Psychiatry, Child and Youth Mental Hlth. Collaborative at the Ctr. for Addiction and Mental Hlth. and The Hosp. for SIck Children, Toronto, ON, Canada; [3]Ctr. for the Developing Brain, Child Mind Inst., New York, NY

Abstract: Introduction In studying the role of biological sex in autism spectrum disorder (ASD), we previously demonstrated coexisting patterns of both shifts towards masculinization and feminization in the intrinsic functional brain properties of males with ASD. Such characterization was not possible in females due to the limited availability of female ASD datasets before. The recent expansion of the Autism Brain Imaging Data Exchange (ABIDE) has overcome this limit allowing for the examination of sex differentiation in the intrinsic brain in females with ASD. **Methods** To explore whether atypical intrinsic brain properties in females with ASD show atypical masculinization and/or feminization, we calculated the spatial overlap between four statistical group difference maps stemming from an autism dataset of females

(ABIDE; ASD vs. neurotypicals [NT]) and an independent normative sample (NT males vs. NT females). The ABIDE sample comprised resting state fMRI (R-fMRI) data from 251 neurotypical females and 111 females with ASD (5.2-46.6 years of age). The normative sex difference sample comprised 471 NT males (NT M) and 357 NT females (NT F; age 8-85 years) selected from the 1000 Functional Connectome Project. We examined a range of R-fMRI measures previously shown to be atypical in ASD and be affected by biological sex. Spatial overlap (conjunction) analyses were performed with logical 'AND' masking of the statistical maps stemming from the ABIDE (ASD vs. NT) and FCP (NT M vs. NT F) comparisons. Two pairs of contrasts represented masculinization (ASD>NT AND NT M> NT F; NT>ASD AND NT F>NT M) and another two represented feminization in ASD (ASD>NT AND NT F>NT M; NT>ASD AND NT M>NT F). Statistical significance was based on the null distribution of random overlap estimated based on 5000 Monte Carlo simulations for any voxel-level thresholds between p=0.05 and 0.0001. **Results** Across all R-fMRI metrics there were consistent, non-random overlaps between regions showing normative sex differences and those showing ASD-NT differences, irrespective of the voxel-level threshold. Similar to previously shown in males with ASD, regions exhibiting masculinized patterns mainly centred around frontal and posterior regions of the default network. Feminization was less pronounced and mainly around somatomotor and ventral attention networks. **Conclusion** These results suggest that atypical intrinsic brain properties of females with ASD partly reflect atypical sexual differentiation. Given the mixed pattern of feminization and masculinization, it is possible that factors involved in typical sex mosaicism are involved in the neurobiology of ASD.

Disclosures: D. Floris: None. **M. Lai:** None. **M.P. Milham:** None. **A. Di Martino:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.21/C11

Topic: A.07. Developmental Disorders

Title: Atypical functional connectome gradients in autism

Authors: *S.-J. HONG[1,2], R. VOS DE WAEL[1,2], S. L. VALK[3], D. S. MARGULIES[4], B. C. BERNHARDT[1,2]
[1]Multimodal Imaging and Connectome Analysis Lab., Montreal, QC, Canada; [2]Montreal Neurolog. Inst., Montreal, QC, Canada; [3]Dept. of Social Neurosci., Max Planck Inst. for Human Cognitive and Brain Sci., Leipzig, Germany; [4]Max Planck Inst. For Human Cognitive and Brain Sci., Leipzig, Germany

Abstract: ***Background.*** Autism spectrum disorder (ASD) is a life-long condition increasingly conceptualized in terms of aberrant brain connectivity [1]. The behavioral symptomatology has

been related to both low- and high-level functions, ranging from atypical sensory processing to disrupted social interactions and language [2]. To study anomalies along the hierarchical organization of the functional connectome in autism, we used a non-linear dimensionality reduction technique to map the principal gradient representing spatial variations in whole-brain connectivity.

Methods. We studied 107 subjects with ASD and 113 typically developing controls from the autism brain imaging exchange (ABIDE) dataset [3]. In all subjects, we mapped resting-state functional MRI signals to corresponding cortical surface models, followed by 3mm surface-based smoothing and the generation of high-dimensional functional connectomes between all pairs of surface-points. Applying a diffusion embedding algorithm [4] to the connectomes identified principal modes/ gradients of connectivity variation across regions. Surface-based linear models compared cortical gradient values between ASD and controls.

Results. Compared to controls, individuals with ASD showed significant alterations in gradient organization (**FIGURE**). Specifically, we observed decreases in gradient strength primarily in bilateral medial and anterior prefrontal cortices - regions co-localizing with core hubs of the default mode network. On the other hand, we observed gradient increases mainly in fronto-parietal and attention networks.

Conclusion. By using a novel connectome gradient mapping, we studied whole-brain functional organization in ASD, and demonstrated an imbalance in the segregation of large-scale functional systems. Further studies will clarify the behavioral relevance of these system-level network reconfigurations.

1. Belmonte MK et al. *J Neurosci* 2004. 24:9228-31; 2. Filipek PA et al. *J Autism Dev Dis* 1999. 29:439-84; 3. Di Martino A et al. *Mol Psych* 2014. 19:659-67; 4. Margulies DS et al. *PNAS* 2016. 113:12574:79

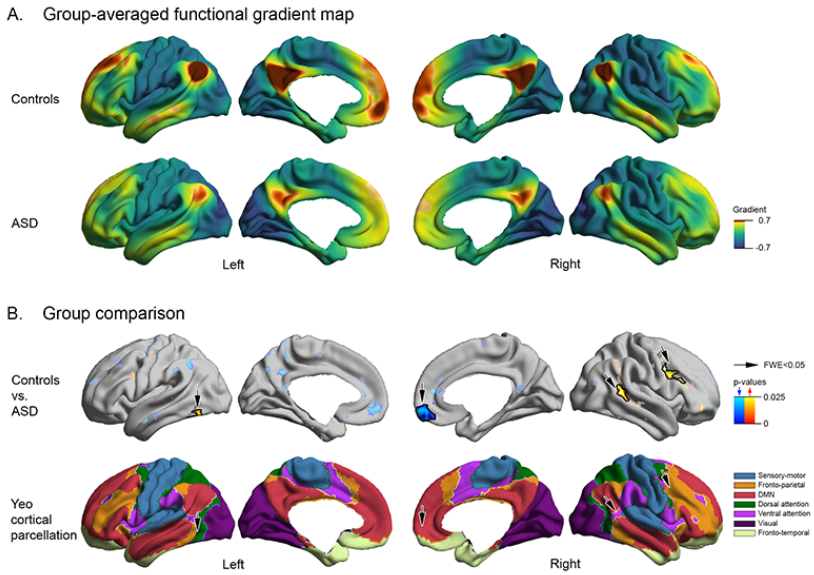


Figure. Altered functional connectome gradients in autism spectrum disorders | A) Group-level functional gradient maps were computed based on averaged similarity matrices of whole-brain connectivity across controls (=113) and individual with autism spectrum disorders (=107). B) A statistical linear model identified group-level differences between controls and ASDs, after correcting for the effects of age, site (*i.e.*, NYU, USM, PITT) and indices of functional signal quality (*i.e.*, Mean framewise displacement and the number of framewise displacement > 0.2mm). The clusters survived for family-wise error controls (random field theory) were highlighted with a sold boundary while the tendency was shown with a semi-transparent color. To help the understanding, an established functional parcellation map (Yeo TT, et al. *J Neurophysiol.* 2011) was shown, indicating specific subnetworks affected in ASD.

Disclosures: **S. Hong:** None. **R. Vos De Wael:** None. **S.L. Valk:** None. **D.S. Margulies:** None. **B.C. Bernhardt:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.22/C12

Topic: A.07. Developmental Disorders

Support: The Hartwell Foundation

Title: Analyzing functional connectivity underlying social/communicative deficits in an autism mouse model

Authors: *B. AN[1], E. THOMSON[2], L. JARED[2], A. H. SUGI[2], A. J. PADILHA[2], C. BAKISKAN[2], M. NICOLELIS[2]
[1]Neurobio., [2]Duke Univ., Durham, NC

Abstract: Autism spectrum disorder (ASD) is a group of neurobiological developmental conditions which are characterized by impaired social/communication abilities and stereotypic behaviors. There has been growing consensus that ASD is associated with aberrant anatomical and functional connectivity throughout the brain, although its neurobiological origins have not been fully elucidated. To examine brain connectivity patterns involved in social/communication impairments of ASD, we employed an ASD mouse model that lacks cereblon gene (CRBN) which has been shown to be involved in intellectual disabilities and speech delay. We examined behaviors of ASD mice in a social context where a mouse was introduced to a stranger mouse, and found that ASD mice displayed less interaction with a stranger mouse and profoundly less vocalization compared to wild type mice. We then investigated whole brain functional connectivity among 16 brain areas implicated in ASD, such as prefrontal cortex, striatum, and amygdala in the social context. We found that signal power throughout the brain of ASD mice was altered: in particular, theta band power was decreased in multiple brain regions, including the striatum and nucleus accumbens (NAc). We further examined pairwise coherence between all recorded regions, and found that theta power coherence was increased between limbic structures of ASD mice. An analysis of functional connectivity, using Granger causality analysis, showed that the amygdala of ASD mice receives stronger inputs from prefrontal cortex, striatum and NAc, suggesting that emotional representation might be altered in ASD mice.

Disclosures: **B. An:** None. **E. Thomson:** None. **L. Jared:** None. **A.H. Sugi:** None. **A.J. Padilha:** None. **C. Bakiskan:** None. **M. Nicolelis:** None.

Poster

369. Autism: Physiology and Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 369.23/C13

Topic: D.07. Vision

Title: Subclinical markers of autism affect low-level visual information processing

Authors: *A. ALINK[1], N. HASSAN[2], F. BOXFORD[2], A. RUNGAPILLAY[2], *S. ONAT[1], C. BÜCHEL[1], I. CHAREST[2]
[1]Dept. for Systems Neurosci., Univ. Med. Ctr. Hamburg-Eppendorf, Hamburg, Germany; [2]Univ. of Birmingham, Birmingham, United Kingdom

Abstract: Previous research suggests that individuals with autism spectrum conditions (ASC) exhibit atypical visual information processing. It is unclear, however, whether visual information processing differs solely between healthy participants and individuals with clinical ASC or whether visual information processing is in general affected by markers of autism. To assess this, we developed a novel experimental paradigm which allows us to obtain a measure of how low-level features of objects contribute to successful object recognition - which we refer to as visual feature diagnosticity (VFD). During the experiment, visual features of cat and dog portrait images were extracted using 1000 Gabor wavelets varying in position, orientation and spatial frequency. On a given trial, participants were presented with a random sample of these features (150) and performed a cat vs. dog classification task. 48 healthy participants took part in this study which included an assessment of their autism-spectrum quotient (AQ) scores - which were all subclinical (below 32). 1000 VFD values for each of the ten stimuli (five cats and five dogs) were obtained for each participant. We determined how VFD values varied as a function of AQ score using a mixed linear model analysis. Because an atypical processing of eyes and of local vs. global information is frequently reported in ASC research we included spatial frequency of visual features and the distance of visual features from the nearest eye as additional predictors in the model. We observed a significant interaction between AQ scores and the distance of a visual feature from the nearest eye. This indicated that a general tendency of visual features closer to the eyes being more important for cat vs. dog discrimination is reduced for individuals with higher AQ scores. Second, our analysis revealed a significant interaction between AQ scores and the spatial frequency of visual features. This interaction highlights a negative relationship between feature spatial frequency and VFD (lower spatial frequencies being more diagnostic) which is more prominent in individuals with higher AQ scores. The first finding is readily reconcilable with previous reports of ASC effects on visual face processing. The second finding appears to be somewhat at odds with previous reports of individuals with ASC being particularly attentive to local details. Importantly, our study reveals a general relationship between markers of autism and visual perception. This opens up the intriguing possibility that also other

personality traits, e.g. extrovertism, are related to individual differences in visual information processing strategies.

Disclosures: **A. Alink:** None. **N. Hassan:** None. **F. Boxford:** None. **A. Rungapillay:** None. **S. Onat:** None. **C. Büchel:** None. **I. Charest:** None.

Poster

370. Rett Syndrome and MECP2

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 370.01/C14

Topic: A.07. Developmental Disorders

Support: KAKENHI 21659252

KAKENHI 23500381

Research grants from The Japan Epilepsy Research Foundation

Grants-in-Aid for Scientific Research on Innovative Areas (Comprehensive Brain Science Network) from the Ministry of Education, Culture, Sports, Science and Technology of Japan

Comprehensive Research on Disability Health and Welfare (Neurological and Muscle diseases) Grant 24-007 of the Ministry of Health, Labor and Welfare of Japan

The Mother and Child Health Foundation Research Grant

Japan Rett Syndrome Support Organization

Title: CDKL5 controls postsynaptic localization of GluN2B-containing NMDA receptors in the hippocampus, and regulates seizure susceptibility, as well as emotional behaviors and memory

Authors: *T. TANAKA[1], K. OKUDA[1], S. KOBAYASHI[2], M. FUKAYA[3], K. TAKAO[4], A. WATANABE[1], T. MURAKAMI[1], M. HAGIWARA[1], S. KOMANO-INOUE[5], H. MANABE[5], M. YAMAGUCHI[5], H. SAKAGAMI[3], T. MIYAKAWA[6], M. MIZUGUCHI[1], T. MANABE[2]
[1]Grad Sch. of Medicine, The Univ. of Tokyo, Tokyo, Japan; [2]Inst. Med. Sci, Univ. Tokyo, Minato-ku, Japan; [3]Dept. of Anatomy, Kitasato Univ. Sch. of Med., Sagamihara, Japan; [4]Life Sci. Res. Ctr., Univ. of Toyama, Toyama-Shi, Japan; [5]Physiol., The Univ. of Tokyo Grad. Sch. of Med., Tokyo, Japan; [6]Fujita Hlth. University, ICMS, Div. Syst. Med., Toyoake, Japan

Abstract: Mutations in the Cyclin-dependent kinase-like 5 (CDKL5) gene cause severe neurodevelopmental disorders accompanied by intractable epilepsies, i.e. West syndrome or atypical Rett syndrome. Here we report generation of the Cdkl5 knockout mouse and show that

CDKL5 controls postsynaptic localization of GluN2B-containing N-methyl-D-aspartate (NMDA) receptors in the hippocampus, and regulates seizure susceptibility, as well as emotional behaviors and memory. Cdkl5 -/Y mice exhibited significantly enhanced anxiety-like behaviors and significant impairment in reference and working memory. Cdkl5 -/Y mice showed normal sensitivity to kainic acid; however, they displayed significant hyperexcitability to NMDA. In concordance with this result, electrophysiological analysis in the hippocampal CA1 region disclosed an increased ratio of NMDA/α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptor-mediated excitatory postsynaptic currents (EPSCs) and a significantly larger decay time constant of NMDA receptor-mediated EPSCs (NMDA-EPSCs) as well as a stronger inhibition of the NMDA-EPSCs by the GluN2B-selective antagonist ifenprodil in Cdkl5 -/Y mice. Subcellular fractionation of the hippocampus from Cdkl5 -/Y mice revealed a significant increase of GluN2B and SAP102 in the PSD (postsynaptic density)-1T fraction, without changes in the S1 (post-nuclear) fraction or mRNA transcripts, indicating a distribution shift of these proteins to the PSD. Immunoelectron microscopic analysis of the hippocampal CA1 region further confirmed postsynaptic overaccumulation of GluN2B and SAP102 in Cdkl5 -/Y mice. Furthermore, ifenprodil abrogated the NMDA-induced hyperexcitability in Cdkl5 -/Y mice, suggesting that upregulation of GluN2B accounts for the enhanced seizure susceptibility. These data indicate that CDKL5 plays an important role in controlling postsynaptic localization of the GluN2B-SAP102 complex in the hippocampus, and that aberrant NMDA receptor-mediated synaptic transmission underlies the pathological mechanisms of the CDKL5 loss-of-function.

Disclosures: **T. Tanaka:** None. **K. Okuda:** None. **S. Kobayashi:** None. **M. Fukaya:** None. **K. Takao:** None. **A. Watanabe:** None. **T. Murakami:** None. **M. Hagiwara:** None. **S. Komano-Inoue:** None. **H. Manabe:** None. **M. Yamaguchi:** None. **H. Sakagami:** None. **T. Miyakawa:** None. **M. Mizuguchi:** None. **T. Manabe:** None.

Poster

370. Rett Syndrome and MECP2

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 370.02/C15

Topic: A.07. Developmental Disorders

Support: CDKL5–16–107–01, UPenn Orphan Disease Center: LouLou Foundation

CDKL5, Caley J. Brown Foundation

Title: Towards epigenomic editing using CRISPR/Cas9 as a putative therapy in CDKL5-deficiency

Authors: *J. A. HALMAI[1], P. DENG[2], H. O'GEEN[2], N. COGGINS[2], J. NOLTA[1], D. SEGAL[2], K. FINK[1]
[1]UC Davis Med. Ctr., Sacramento, CA; [2]Genome Ctr., UC Davis, Davis, CA

Abstract: Neurological diseases are a heterogeneous group of disorders caused by alterations in nervous system function and due to technological advances over the last decade; many of these disorders can be attributed to genetic factors such as chromosomal aberrations or gene mutations. Early onset epileptic encephalopathies are among the most devastating conditions among pediatric populations. Our research is focused on methods to reactivate the healthy *CDKL5* gene on the silenced X-chromosome in CDKL5-deficient patient cells and cell lines. Our team has validated several novel genomic targets that can regulate *CDKL5* expression in human neuronal cells. This technique utilizes CRISPR guide RNAs (gRNAs) with deactivated Cas9 fused VP64 (dCas9-VP64) targeted to regulatory domains in the *CDKL5* gene and results in significant upregulation of the gene when transfected with a pooled group of gRNAs. We believe this to be the first description of a true regulatory domain in the *CDKL5* promoter. We have performed chromatin immunoprecipitation qPCR to examine the recruitment of sustainable and durable changes to the epigenetic landscape following co-transfection or transduction with the same pooled gRNAs paired with dCas9-VP64 into human neurons. We have also expanded the library of effector domains that can be paired with our pooled gRNAs and successfully drive gene activation. These novel effector domains may be able to provide a more durable and sustainable reactivation of the silenced X-chromosome in a therapeutic setting. In addition, we have designed, cloned, and validated gRNAs for both the murine and rat *CDKL5* gene to be able to perform gene activation studies in transgenic rodent models of CDKL5-deficiency. Our group specializes in the creation of TALE and CRISPR to target and permanently reverse aberrant genetic mutations that contribute to disease. To our knowledge we are the first to demonstrate significant up-regulation of endogenous *CDKL5* using DNA binding domains. This approach holds great potential for children suffering from CDKL5-deficiency.

Disclosures: J.A. Halmai: None. **P. Deng:** None. **H. O'Geen:** None. **N. Coggins:** None. **J. Nolta:** None. **D. Segal:** None. **K. Fink:** None.

Poster

370. Rett Syndrome and MECP2

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 370.03/C16

Topic: A.07. Developmental Disorders

Support: 91332203

Title: MECP2 mutation causes abnormal effect of D2R agonist on locomotor and thermoregulation in mice

Authors: *B. LU, Z. XIONG
Lab. of Neurobio. of Diease, Inst. of Neurosci., Shanghai City, China

Abstract: X-linked *MECP2* gene plays an essential role in development of central neuron system. Its mutation is the main cause of neurodevelopmental disorder Rett syndrome. Mutations in *MECP2* cause motor skill impairment as well as kinds of neuropsychiatric syndrome in humans. The mechanisms mediating these features, however, remain unclear. Here we reported that Mecp2 is involved in the functional maturation of dopamine D2-like receptors in motor and thermoregulation circuits. Our data showed that Mecp2 mutant mice displayed abnormal locomotion in response to administration of D2-like receptor agonist (quinpirole) in novel environment. We also found that hypothermia effect of quinpirole was reduced in Mecp2 mutant mice compared to their wild type littermates. In addition we found that the phenotype of abnormal locomotion and thermoregulation in response to quinpirole just existed in adult mice but not in pup (P18-P20). The present study indicated the important role of Mecp2 in postnatal maturation of D2R-associated circuits in locomotor and thermoregulation, which may provide insights into the cellular and circuitry mechanisms underlying the pathogenesis of Rett syndrome.

Disclosures: B. Lu: None. **Z. Xiong:** None.

Poster

370. Rett Syndrome and MECP2

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 370.04/C17

Topic: A.07. Developmental Disorders

Support: Center for Nanoscale Microscopy and Molecular Physiology of the Brain (CNMPB)

Title: Neuronal redox imbalance in Rett syndrome: a key player in neuronal network dysfunction and altered neurotransmitter responsiveness?

Authors: K. CAN, K. FESTERLING, S. KÜGLER, *M. MUELLER
Univ. Goettingen, Goettingen, Germany, Germany

Abstract: Rett syndrome (RTT) is a neurodevelopmental disorder, affecting almost exclusively girls. Its main genetic cause are *de novo* mutations in the methyl-CpG binding protein 2 gene (*MECP2*). After a short and apparently normal development, a RTT child enters a developmental stagnation, followed by neuronal and autonomic dysfunction, which manifest as mental retardation, erratic breathing, epilepsy, loss of speech, stereotypical hand movements and motor disturbances. Furthermore, RTT is associated with early mitochondrial dysfunction and systemic oxidative stress. We showed previously that mitochondria of MeCP2-deficient (*Mecp2*$^{-/y}$) mouse

hippocampus operate at increased turnover rates. In concert with impaired cell-endogenous scavenging systems, this culminates in cellular redox dysregulation. To uncover molecular details of this redox impairment, we extended optical redox imaging specifically to neurons. Taking advantage of viral vectors (AAV-6), we expressed genetically-encoded redox sensors in either cytosol (roGFPc) or mitochondrial matrix (roGFPm). In dissociated cell cultures, stimulation by glutamate, dopamine, serotonin, and norepinephrine consistently evoked intensified oxidizing shifts in the cytosol of *Mecp2*$^{-/y}$ neurons, suggesting that even physiological stimuli such as neurotransmitters are sufficient to provoke overshooting redox responses. In mitochondria, neurotransmitter-evoked redox changes were more moderate than in cytosol, and genotypic differences between WT and *Mecp2*$^{-/y}$ neurons were less pronounced. Cellular Ca^{2+} overload due to massive Ca^{2+} influx can be excluded as a primary cause for the neurotransmitter-induced redox responses and their overshooting characteristics in *Mecp2*$^{-/y}$ neurons. Instead, by pharmacological intervention we confirmed a leading role of NADPH- and xanthine oxidases in cytosolic ROS production. This identifies the cytosol as an important ROS source, but also as an unsufficiently redox-buffered compartment. Interestingly, the very route of activation of these oxidases apparently differs among *Mecp2*$^{-/y}$ and WT neurons. All of these changes were evident already in neonatal and presymptomatic mice. Therefore, it will be crucial to map cellular redox conditions as RTT progresses in severity. A crucial tool for this future endeavor will be our recently generated redox-indicator mice, which are currently crossbred with Rett mice. Only then, it will become evident to what degree early neuronal redox alterations contribute to neuronal network dysfunction and promote the further progression of RTT.

Disclosures: K. Can: None. **K. Festerling:** None. **S. Kügler:** None. **M. Mueller:** None.

Poster

370. Rett Syndrome and MECP2

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 370.05/C18

Topic: A.07. Developmental Disorders

Support: Jerome Lejeune Foundation

ProRett Italia

Title: Defective mechanisms of corticogenesis in MeCP2 null cerebral cortexes

Authors: *F. BEDOGNI[1], C. COBOLLI GIGLI[1], L. SCARAMUZZA[1], A. MORIN[1], R. ROSSI[2], N. LANDSBERGER[3]
[1]San Raffaele Hosp., Milano, Italy; [2]Inst. Nazionale Genetica Molecolare, Milan, Italy; [3]Univ. of Milan, Milan, Italy

Abstract: The X-linked Methyl-CpG-Binding Protein 2 (MeCP2) gene encodes for a multifunctional protein ubiquitously expressed from developmental stages to adulthood. Mutations in *MECP2* are linked to Rett syndrome (RTT), the most common genetic cause of severe intellectual disability in females. Although MECP2 plays a crucial role in the maintenance of proper neuronal functionalities, several evidences now suggest that early signs of the pathology can be observed (in both humans and animal models) long before the typical RTT symptoms become overt. We focused on the development of the embryonic cerebral cortex of *Mecp2* null animals, as our published transcriptional analyses suggest that the mechanisms of embryonic corticogenesis are delayed. Based on this observation, we assessed the dynamics of neuronal differentiation in embryonic and early postnatal cortical tissues. Our data show that the expression of transcripts that are typical of neuronal progenitors is retained by null newborn neurons, while the level of transcripts expressed by maturing or fully functional neurons are reduced in null cortexes. Altogether, our data suggest that already during embryonic and early post-natal life lack of Mecp2 affects the ability of newborn neurons to drop the transcription of genes that are outdated (like those defining cortical progenitors) while acquiring new, more refined, postmitotic identities. We believe this evidence demonstrates that the transcriptional noise typically affecting adult *Mecp2* null tissues is a feature already displayed at an early stage of differentiation. The impairments displayed by adult RTT animal models can thus be considered the worsening of a condition that is already generated during embryogenesis.

Disclosures: F. Bedogni: None. **C. Cobolli Gigli:** None. **L. Scaramuzza:** None. **A. Morin:** None. **R. Rossi:** None. **N. Landsberger:** None.

Poster

370. Rett Syndrome and MECP2

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 370.06/C19

Topic: A.07. Developmental Disorders

Support: Parent association l'albero di Greta

Pennsylvania Orphan Disease Center and London-based LouLou Foundation

Title: Innovative pregnenolone-based therapeutic approaches for neurological disorders linked to mutations in X-linked Cyclin dependent kinase-like 5 (CDKL5)

Authors: *I. BARBIERO[1], D. PERONI[1], M. TRAMARIN[1], L. RUSCONI[1], P. MOTTA[1], P. SINISCALCHI[1], N. LANDSBERGER[2], M. BIANCHI[3], C. KILSTRUP-NIELSEN[1]
[1]DBSV, Univ. of Insubria, Busto Arsizio, Italy; [2]Dept. of Med. Biotech. and Translational Medicine,, Univ. of Milan, Milan, Italy; [3]Transpharmation Ireland Limited, Dublin, Ireland

Abstract: Mutations in the X-linked cyclin-dependent kinase-like 5 (CDKL5) gene cause neurologic disorders characterized by the early onset of seizures, intellectual disability, impairment of gross motor skills, and sleep disturbances. CDKL5 is a serine/threonine kinase the molecular network of which is not yet fully understood. Loss of CDKL5 both in vitro and in vivo leads to altered neuronal morphology including defective polarization, dendritic arborization and spine morphology suggesting a link between CDKL5 and the cytoskeleton.
Recently, we identified IQGAP1 as a novel interactor of CDKL5. IQGAP1 is a scaffold protein that regulates cytoskeleton dynamics by linking the actin and microtubule (MT) networks through its association with Rac1 and the MT plus end binding protein (+TIP) CLIP170. CDKL5 deficiency interferes with the formation of the IQGAP1/CLIP170/Rac1 ternary complex with repercussions on the MT association of CLIP170. These findings allowed us to recognize the neurosteroid pregnenolone (P5) as an innovative candidate drug for CDKL5-related disorders. Indeed, CLIP170 acts as an intracellular receptor for P5 that promotes microtubule dynamics by blocking CLIP170 in its active conformation. Accordingly, we find that treatment of CDKL5 deficient cells with P5 is capable of restoring the MT association of CLIP170 and a proper neuronal morphology.
Although P5 has well-known positive effects on cognition and neuronal morphology in animal models and has proven to be well tolerated in clinical tests, there is some concern about its clinical application due to the possible side effects of its downstream metabolites. In such a scenario, synthetic P5 derivatives like pregnenolone-methyl-ether, PME, which maintain the biological action but cannot be converted into other steroids, is therefore of high clinical relevance. These molecules are currently considered innovative drug candidates for psychiatric and neurodegenerative disorders. Here we present data showing that a range of morphological and molecular defects in primary neuronal cultures from Cdkl5-null mice can be restored upon treatment with either P5 or PME suggesting their potential therapeutic relevance for CDKL5-related disorders.

Disclosures: I. Barbiero: None. **D. Peroni:** None. **M. Tramarin:** None. **L. Rusconi:** None. **P. Motta:** None. **P. Siniscalchi:** None. **N. Landsberger:** None. **M. Bianchi:** None. **C. Kilstrup-Nielsen:** None.

Poster

370. Rett Syndrome and MECP2

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 370.07/C20

Topic: A.07. Developmental Disorders

Title: Efficacy of ANAVEX 2-73, a Sigma-1 receptor agonist, in the MECP2 mouse model of Rett syndrome

Authors: *J. S. SPROUSE, N. REBOWE, D. KLAMER, C. MISSLING
Anavex Life Sci., New York, NY

Abstract: BACKGROUND: The Sigma-1 receptor (σ1R) is an intracellular chaperone protein located at the endoplasmic reticulum - mitochondria interface with important roles in inter-organelle communication and the cellular response to stress. ANAVEX 2-73 (AV2-73) is a σ1R agonist that has previously demonstrated favorable safety, bioavailability, and tolerability in Phase 1/2 clinical trials. Data from the ongoing Phase 2a study in Alzheimer's disease patients demonstrate signs of dose-dependent cognitive improvement. Given the reported ability of the σ1R to restore cellular functionality, neurodevelopmental disorders may respond to the activation of σ1R in a disease-modifying manner. One such disorder is Rett syndrome and the MECP2 HET mouse is a well-characterized model with a behavioral profile that mimics many aspects of the clinical picture.
METHODS: Female MECP2 HET and wild type (WT) mice were used throughout (N=19-20 per treatment arm). Chronic daily dosing of AV2-73 (10 or 30 mg/kg/day PO) starting at 5.5 weeks of age was conducted throughout a 12-week period of testing. Behavioral paradigms measured different aspects of motor coordination, reflex reactivity, and species-specific behavior (clasping). In a separate study, 4 weeks of daily dosing starting at 6.5 months of age was followed by optokinetic analysis of relative visual acuity and changes in respiration by whole body plethysmography. Significance at $p<0.05$ was determined by ANOVA and post-hoc comparisons.
RESULTS / DISCUSSION: In the younger cohort of mice, chronic dosing with AV2-73 significantly improved performance of the MECP2 HETs in different motor and gait paradigms, and reduced clasping behavior to WT levels. Among the older cohort, relative visual acuity in AV2-73-treated HET mice was returned to WT levels at the slower rotating speed. A reduction in apnea counts (~35%) relative to WT levels was observed in HETs receiving AV2-73.
CONCLUSIONS: AV2-73 significantly improves an array of behavioral phenotypes in the Rett syndrome mouse model in a dose-related manner. Based on these data, Anavex Life Sciences will start a U.S. multicenter Phase 2 clinical trial of AV2-73 for the treatment of Rett syndrome with the support of Rettsyndrome.org.

Disclosures: **J.S. Sprouse:** F. Consulting Fees (e.g., advisory boards); Anavex Life Sciences. **N. Rebowe:** A. Employment/Salary (full or part-time):; Anavex Life Sciences. **D. Klamer:** A. Employment/Salary (full or part-time):; Anavex Life Sciences. **C. Missling:** A. Employment/Salary (full or part-time):; Anavex Life Sciences.

Poster

370. Rett Syndrome and MECP2

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 370.08/C21

Topic: A.07. Developmental Disorders

Support: NIH-NINDS

HFSP-CDF

Title: Impact of arousal on visual processing in Rett syndrome

Authors: *P. ARTONI[1], G. A. EWALL[2], G. RANKIN[1], C. CHEN[1], T. K. HENSCH[2], M. FAGIOLINI[1]
[1]F. M. Kirby Neurobio. Ctr., Boston Children's Hospital, Harvard Med. Sch., Boston, MA; [2]Mol. and Cell. Biol., Harvard Univ., Cambridge, MA

Abstract: Proper sensory processing is actively modulated by arousal states. Growing evidence implicates abnormal visual and auditory processing in Rett Syndrome (RTT), a neurodevelopmental disorder caused by *de novo* mutation of the X-linked MECP2 gene. RTT is characterized by an initial period of apparent normal development followed by stagnation and subsequent regression of sensory processing, cognition and motor function. Onset of epilepsy and anxiety-like behavior often occur later. How neuronal cortical circuits are disrupted during disorder progression and whether arousal states contribute further to such disruptions is still largely unknown. Here, we performed longitudinal *in vivo* 2-photon imaging of Ca^{++} transients in visual cortical circuits of freely moving $MeCP2^{stop/y}$ and wild-type (WT) littermate mice while tracking motor activity, pupil diameter and gaze. At postnatal day P30 (before regression begins and RTT-like phenotype increases), animals were injected with AAV9-GCaMP6s into visual cortex and implanted with a cranial window and head post. After a 2-week habituation to the running ball set-up and head restraint, mice were imaged from the binocular area of visual cortex during their active phase (nocturnal). We alternately measured Ca^{++} transients of spontaneous or evoked activity in layer 2/3 cells in the absence of stimulation (grey screen) or in response to visual input (drifting gratings of different spatial frequencies, 100% contrast, 4Hz temporal frequency), respectively. Mice underwent multiple imaging sessions over the course of the disease progression. We sorted the data into two groups: active regression (AR, P45- P80) and late regression (LR, P80 - P110). We found that the spontaneous activity in MeCP2-deficient mice was significantly lower both in AR and in LR stages compared to WT (WT: N=5 mice, n=489 cells; AR: N=4, n=133; LR: N=5, n=149, respectively). Receptive field properties such as direction and orientation selectivity or spatial resolution of individual cells progressively deteriorated from AR to LR stages. In contrast to the increase displayed in WT mice, there was no significant positive modulation of spontaneous activity levels across arousal states in the mutant mice. Moreover, visual acuity and response reliability were detrimentally impacted by the high arousal state. Overall, our data suggest that in mice lacking MeCP2, the decline of visual cortical processing correlates with the onset of the disorder, and it is further disrupted by heightened arousal states.

Disclosures: **P. Artoni:** None. **G.A. Ewall:** None. **G. Rankin:** None. **C. Chen:** None. **T.K. Hensch:** None. **M. Fagiolini:** None.

Poster

370. Rett Syndrome and MECP2

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 370.09/C22

Topic: A.07. Developmental Disorders

Support: NIH/OD DP5OD009134

NIH/NICHD U54HD083092

NIH/NICHD R01HD083181

The Stedman West Foundation

Texas Children's Hospital

Title: Replication and comprehensive evaluation of neurobehavioral outcomes in a rat model of Mecp2

Authors: *S. VEERARAGAVAN, S. G. HUANG, C. S. WARD, R. C. SAMACO
Mol. and Human Genet., Baylor Col. of Med., Houston, TX

Abstract: Rett syndrome (RTT) is an X-linked disorder caused by loss-of-function mutations in gene encoding the transcriptional modulator Methyl-CpG-Binding Protein 2 (MeCP2). Typical RTT occurs in girls and women and is characterized by a brief period of normal development followed by regression of acquired hand skills and language, and the onset of anxiety, autistic features, seizures and autonomic dysfunction. However, rare cases of boys that possess similar mutations in *MECP2* present with severe congenital encephalopathy accompanied with severe developmental delay, hypotonia, seizures and severe autonomic dysfunction. Furthermore, these affected boys do not survive past early childhood. Recent studies by our group and others have demonstrated the value of a novel *Mecp2* rat model for both mechanistic and translational studies for MeCP2-related disorders. However, to avoid experimental pitfalls such as issues of reproducibility and reliability, especially for neurobehavioral outcomes commonly studied in mouse models of ASD/IDD including *Mecp2* mouse models, we set out to evaluate male rats completely lacking MeCP2, choosing to confirm and expand upon the limited behavioral data so far reported in *Mecp2* male rats. Two of the three reports on the *Mecp2* rat model examined spontaneous exploratory activity, with one study conducting additional tests for grip strength and sociability, and another study evaluating gait. Consistent with previous findings, we observed a significant decrease in overall spontaneous exploratory activity in *Mecp2* male rats. Our additional behavioral studies also uncovered features not previously reported including decreased anxiety-like behavior, enhanced self-grooming, evidence for decreased aspects of juvenile play, and sensorimotor gating abnormalities. We also tested aversive and episodic-like learning and

memory in the *Mecp2* male rats and observed no significant differences across genotypes. This replication study provides a timely assessment of the robustness and reproducibility of current findings, as the field rapidly pushes forward in incorporating this novel rodent model as a key component in studies of biological mechanisms and potential therapies for MeCP2-related disorders.

Disclosures: S. veeraragavan: None. **S.G. Huang:** None. **C.S. Ward:** None. **R.C. Samaco:** None.

Poster

370. Rett Syndrome and MECP2

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 370.10/C23

Topic: A.07. Developmental Disorders

Support: R21NS10085

Title: Microglia phenotype varies regionally in in a Mecp2-heterozygous model of Rett Syndrome

Authors: *C. O'FERRALL[1], A. FOWLER[3], E. S. SMITH[1], M. S. LANGE[3], G. E. HOFFMAN[3], M. E. BLUE[4], S. KANNAN[2]
[1]Critical Care Med., [2]Anesthesiol. and Critical Care Med., Johns Hopkins Univ., Baltimore, MD; [3]Biol., Morgan State Univ., Baltimore, MD; [4]Hugo W Moser Res. Inst., Kennedy Krieger Inst., Baltimore, MD

Abstract: Microglia have been implicated in the neuropathology observed in mouse models of Rett Syndrome (RTT). However, many of the impairments in microglia function seem to be independent of MeCP2 presence within the cell, as others have shown expression of MeCP2 in *Mecp2*-null mice is insufficient in repairing behavioral deficits. Much less is known about the role of microglia in *Mecp2*-heterozygous female mice, which match the genotype of human RTT. The current study tested the hypothesis that microglia phenotype/profile varies across affected brain areas in a mouse model of RTT. Brain tissue from 6-12 month old *Mecp2*-heterozygous female mice were fixed, preserved and processed for Iba-1, CD68 (marker of phagocytosis) and ferritin (protein integral in iron storage) immunocytochemistry. The distribution of microglia was examined in hippocampus, striatum, arcuate nucleus, amygdala, and prefrontal cortex. Different morphologic parameters of microglial cells including cell size, process number and process length were quantified using Neurolucida imaging software. The morphological analyses demonstrated a distinct M1/pro-inflammatory microglial profile (larger somas, fewer, shorter processes) in the arcuate nucleus that was particularly marked in overweight *Mecp2*-heterozygous mice. Patterns of CD68 and ferritin immunostaining were

distinctive in hippocampus but not in other brain regions studied. The results indicate region-specific differences in microglia phenotype/profile in *Mecp2*-heterozygous mice. Ongoing studies are characterizing the interactions between microglia and the larger neuronal environment within these brain regions to further elucidate the contributions of microglia to the neuropathology in RTT.

Disclosures: C. O'Ferrall: None. **A. Fowler:** None. **E.S. Smith:** None. **M.S. Lange:** None. **G.E. Hoffman:** None. **M.E. Blue:** None. **S. Kannan:** None.

Poster

370. Rett Syndrome and MECP2

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 370.11/C24

Topic: A.07. Developmental Disorders

Support: NIH F31 Grant NS098574-02

NIH Grant NS057398

Title: Ketamine treatment rapidly reverses cortical synaptic spine deficits in a mouse model of Rett syndrome

Authors: C. J. HOWELL, 44120[1], S. U. LAD[2], F. E. ABOUELSOUD[2], *D. M. KATZ[3]
[1]Neurosciences, [3]Dept Neurosciences, [2]Case Western Reserve Univ., Cleveland, OH

Abstract: Rett syndrome (RTT), a severe disorder caused by loss-of-function mutations in the *MECP2* gene, is characterized by neurological regression leading to deficits in motor, respiratory and autonomic control and cognitive function. Similar to other autism spectrum disorders, RTT is characterized by abnormal synaptic connectivity, including reduced density and maturity of cortical dendritic spines. In the medial prefrontal cortex (mPFC), a region critical for behaviors disrupted in RTT, spine deficits on Layer 5 pyramidal neurons are associated with decreased excitatory synaptic drive and reduced neuronal activity (Sceniak et al., 2015; Kron et al., 2012). Moreover, hypoactivity of mPFC pyramidal neurons appears to underlie or contribute to neurological deficits in *Mecp2* mutant mice (Howell, et al., 2016), suggesting that restoration of excitatory connectivity in the mPFC could be of therapeutic benefit in RTT. Therefore, the present study was designed to test the reversibility of spine defects in *Mecp2* null (Null) mice in response to ketamine, an NMDA antagonist which promotes dendritic spine growth in rodent models of depression (Li et al., 2010). We also examined the impact of ketamine on signaling mechanisms that are associated with spine growth and are in deficit in the Null brain (Ricciardi et al., 2011), including phosphorylation of the ribosomal protein S6, a target of mTOR (*mechanistic target of rapamycin*). To approach these issues we compared phospho-S6 signaling

and dendritic spine density in the mPFC of symptomatic Null and wildtype (WT) mice treated with a single sub-anesthetic dose of ketamine. To visualize dendritic spines, we used Thy1-GFPm *Mecp2* Null or WT mice in which GFP is expressed in a subset of layer V pyramidal neurons. 24 hours after treatment, the density of spines on oblique dendrites was significantly increased to WT levels in ketamine-treated *Mecp2* mutants. This rescue was due in part to significant increases in the density of stubby and thin spines. To determine if these effects of ketamine were associated with changes in S6 signaling in the mPFC, we compared levels of total and phospho-S6 in saline- and ketamine-treated Wt and Null mice using Western blot. Within 1 hour of treatment, phospho-S6 levels were significantly increased to WT levels in ketamine-treated *Mecp2* mutants. Thus, even in the absence of MeCP2, spine deficits in cortical pyramidal neurons can be rapidly reversed with a single sub-anesthetic dose of ketamine. These data support the potential of low-dose ketamine treatment to improve neurological function in RTT by promoting restoration of excitatory synaptic connectivity.

Disclosures: C.J. Howell: None. **S.U. Lad:** None. **F.E. Abouelsoud:** None. **D.M. Katz:** Other; Founding advisor to ArRETT Neuroscience, a company focused on advancing treatments for patients with Rett syndrome..

Poster

370. Rett Syndrome and MECP2

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 370.12/C25

Topic: A.07. Developmental Disorders

Support: GSK-CIHR chair

The Richard and Edith Strauss post-doctoral Fellowships

Title: Methyl-CpG binding domain protein 2 is a master regulator of neuronal gene pathways and behavior

Authors: *E. M. LAX, S. DO-CARMO, Y. ENUKA, N. MAHMOOD, S. RABBANI, C. A. CUELLO, Y. YARDEN, M. SZYF
Dept. of Pharmacol. and Therapeutics, Fac. of Med., McGill Univ., Montreal, QC, Canada

Abstract: DNA methylation is a major epigenetic modification that alter gene expression. Methyl-CpG-binding domain (MBD) proteins recognize and bind methylated DNA. Several MBDs are involved in neurodevelopment and maintenance of brain functions. However, not much is known about the role of Methyl-CpG-binding domain 2 (Mbd2) in the brain. We found MBD2 knockout mice show deficits in cognitive, social and emotional functions. Chromatin immunoprecipitation (ChIP-seq) revealed that Mbd2 binds to regulatory DNA regions of

neuronal genes in the Hippocampus. Loss of Mbd2 alters the expression of hundreds of genes with a robust down-regulation of neuronal gene pathways. Further, a genome-wide DNA methylation analysis found an altered DNA methylation pattern in regulatory DNA regions upon loss of Mbd2. Taken together, our data suggest Mbd2 regulates hippocampal DNA methylation landscape to control neuronal gene expression and behavior.

Disclosures: E.M. Lax: None. **S. Do-Carmo:** None. **Y. Enuka:** None. **N. Mahmood:** None. **S. Rabbani:** None. **C.A. Cuello:** None. **Y. Yarden:** None. **M. Szyf:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.01/C26

Topic: A.09. Adolescent Development

Support: NIMH MH107703

NIMH MH107235

NIMH MH089983

NIMH MH089924

Dowshen Program for Neuroscience MH102609

Title: Data-driven assessment of structural image quality

Authors: *A. F. ROSEN[1,2], D. R. ROALF[2], K. RUPAREL[2], J. BLAKE[2], K. SEELAUS[2], L. VILLA[2], P. A. COOK[3], C. DAVATZIKOS[4,3], M. A. ELLIOT[3], A. GARCIA DE LA GARZA[2], E. D. GENNATAS[2], M. QUARMLEY[2], J. SCHMITT[3,2], R. T. SHINOHARA[5], M. TISDALL[3], R. CRADDOCK[6,7], R. E. GUR[2,3], R. C. GUR[2,3], T. D. SATTERTHWAITE[2]
[1]Psychiatry, [2]Neuropsychiatry, [3]Radiology, [4]Electrical and Systems Engin., [5]Biostatistics, Epidemiology, and Informatics, Univ. of Pennsylvania, Philadelphia, PA; [6]Ctr. for Developing Brain, Child Mind Inst., New York, NY; [7]Computat. Neuroimaging Lab., Nathan S. Kline Inst., New York, NY

Abstract: ABSTRACT
Data quality is increasingly recognized as one of the most important confounders in brain imaging research. It is particularly important for studies of brain development, where age is negatively related to in-scanner motion and data quality. Prior work has demonstrated that in-scanner head motion biases estimates of structural neuroimaging measures. Yet, objective measures of data quality are not available for most structural brain images. Here we sought to

identify reliable, quantitative measures of data quality for T1-weighted volumes, describe how such measures of quality relate to common measures of brain structure, and delineate how this in turn may bias inference regarding brain development in youth. Three expert raters provided manual ratings for 1601 T1-weighted volumes acquired as part of the Philadelphia Neurodevelopmental Cohort. Manual ratings were compared to automated quality measures, which derived measures from the Preprocessed Connectomes Project's Quality Assurance Protocol (QAP). Generalized linear mixed-effects models using the automated quality measures were constructed in a training sample (n = 1067) to: 1) identify unusable images with significant artifacts, and 2) quantify subtle artifacts in usable images. These models were then tested in an independent validation dataset (n = 534). Results reveal that unusable images can be detected with a high degree of accuracy: a model including background kurtosis and skewness achieved an AUC of 0.95 in the training dataset and 0.94 in the independent validation dataset. While identification of subtle artifact was more challenging, an 8-parameter model achieved an AUC of 0.80 in the training dataset, and 0.92 in the validation dataset. Notably, quantitative measures of image quality were related to cortical thickness and gray matter density; measures of cortical volume were less affected by artifact. Furthermore, these quantitative measures of image quality explained more variance structural biomarkers then estimates of motion derived from other imaging sequences acquired during the same protocol. Finally, data quality significantly altered estimated structural brain maturation occurring during adolescent development. Taken together, these results indicate that reliable measures of data quality can be automatically derived from T1-weighted volumes, and that failing to control for data quality can systematically bias the results of studies of brain development.

Disclosures: **A.F. Rosen:** None. **D.R. Roalf:** None. **K. Ruparel:** None. **J. Blake:** None. **K. Seelaus:** None. **L. Villa:** None. **P.A. Cook:** None. **C. Davatzikos:** None. **M.A. Elliot:** None. **A. Garcia de La Garza:** None. **E.D. Gennatas:** None. **M. Quarmley:** None. **J. Schmitt:** None. **R.T. Shinohara:** None. **M. Tisdall:** None. **R. Craddock:** None. **R.E. Gur:** None. **R.C. Gur:** None. **T.D. Satterthwaite:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.02/C27

Topic: A.09. Adolescent Development

Title: In-scanner head motion systematically impacts estimates of structural connectivity: Implications for studies of structural brain network development

Authors: ***G. L. BAUM**[1], D. R. ROALF[1], P. A. COOK[3], R. CIRIC[1], A. ROSEN[1], M. A. ELLIOT[3], P. RUPERT[1], M. QUARMLEY[1], K. RUPAREL[1], R. C. GUR[1], R. E. GUR[1], D. S.

BASSETT[2], T. D. SATTERTHWAITE[1]
[1]Neuropsychiatry, [2]Bioengineering, Univ. of Pennsylvania, Philadelphia, PA; [3]Radiology, Univ. of Penssylvania, Philadelphia, PA

Abstract: Connectomics provides a powerful analytical framework for delineating brain development during adolescence. One challenge in MRI studies of brain network development in childhood and adolescence is data quality: younger subjects tend to move more during image acquisition, and differences in head motion have been shown to significantly bias developmental patterns of functional connectivity. Despite this fact, the impact of motion artifact on structural connectivity networks remains poorly characterized. Here, we evaluated the impact of head motion on estimates of structural connectivity using a sample of 949 participants (ages 8-23 years) who passed a rigorous quality assessment protocol for diffusion tensor imaging (DTI) acquired as part of the Philadelphia Neurodevelopmental Cohort. Structural connectivity was measured after constructing brain networks using both deterministic and probabilistic tractography methods, while head motion was estimated by the mean relative displacement between non-diffusion-weighted volumes in the DTI acquisition. We hypothesized that subtle variation in head motion would bias estimates of structural connectivity in a distance-dependent manner, as observed in previous studies of functional connectivity.
Even following rigorous manual quality assurance and retrospective head motion correction with FSL's *eddy*, in-scanner motion impacted the strength of structural connectivity in a distance-dependent manner (permutation-based $p<0.0001$): specifically, increased head motion was associated with reduced estimates of structural connectivity for long-range, consistent connections with high mean fractional anisotropy. In contrast, motion inflated estimates of structural connectivity for short-range, inconsistent connections with low mean fractional anisotropy. Head motion also systematically impacted measures of structural network topology derived from both deterministic and probabilistic tractography, including network density (r=-0.43, $p<10^{-10}$), global efficiency (r=-0.34, $p<10^{-10}$), and modularity (r=-0.16, p=1.2 x 10^{-5}). Controlling for the contributions of subject motion also mitigated estimated age effects on structural connectivity, suggesting that observed developmental changes in structural brain networks may be inflated by age-related differences in head motion. Taken together, these data delineate the systematic impact of head motion on structural connectivity using a variety of brain network construction methods, and provide a critical framework for quantifying and mitigating motion-related confounds in studies of structural brain development.

Disclosures: G.L. Baum: None. **D.R. Roalf:** None. **P.A. Cook:** None. **R. Ciric:** None. **A. Rosen:** None. **M.A. Elliot:** None. **P. Rupert:** None. **M. Quarmley:** None. **K. Ruparel:** None. **R.C. Gur:** None. **R.E. Gur:** None. **D.S. Bassett:** None. **T.D. Satterthwaite:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.03/C28

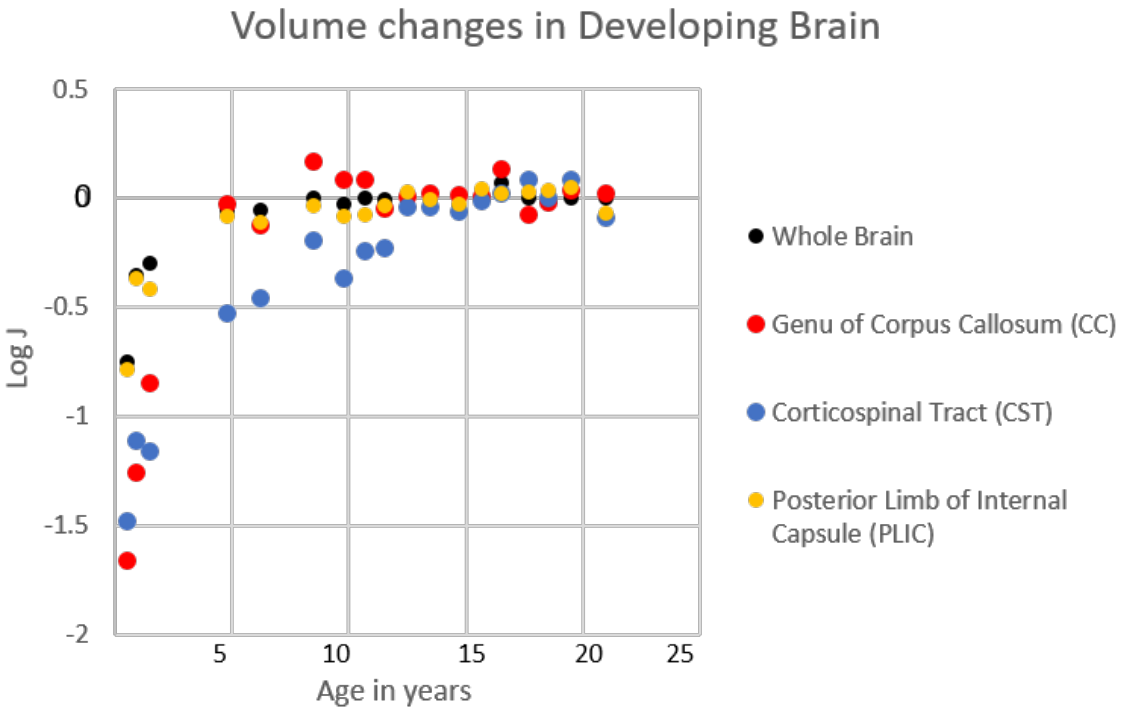
Topic: A.09. Adolescent Development

Support: NIH Intramural program

Title: Investigating postnatal human brain development using diffusion tensor based morphometry (DTBM)

Authors: A. NAYAK, M. IRFANOGLU, N. SADEGHI, *C. PIERPAOLI
Quantitative Med. Imaging Section, NIBIB, NIH, Bethesda, MD

Abstract: MRI can be used to assess postnatal brain development. Tensor Based Morphometry (TBM), performed from T1weighted images (T1WIs), is used to measure local differences in volume when a brain is warped to another brain or a template[1]. However, T1W signal is homogenous in white matter and can not be used to detect volume changes in specific white matter pathways. Diffusion Tensor Imaging (DTI) allows the identification of specific white matter pathways that appear homogeneous on T1WIs. The novel approach of this work is to assess morphometric changes in specific white matter pathways performing TBM with DTI. We name this approach: DTBM.
We used DTI data from182 subjects in the NIH MRI Normal Brain Development public database[2] to create 19 age specific DTI average brain templates. The templates were created by co-registering data of individual subjects in each age range using the recently proposed Diffeomorphic Registration for Tensor Accurate Alignment of Anatomical Structures (DR-TAMAS) algorithm[3]. An adult template for the population was created from subjects in the 18-21yr old age group. Each age-specific template was then registered to the adult template using DR-TAMAS. The voxelwise Log of the determinant of the Jacobian (Log J) was computed to quantify local volume differences with respect to the adult template. ROIs were drawn on several white matter pathways on the adult template and average Log J values were computed for each age specific template.
In the figure it is interesting to note that different white matter pathways have very different volume growth trajectories (VGT). for example, PLIC VGT is aligned with the overall brain VGT. CST and CC have much larger volume changes, with very different rate of change.
In conclusion, our resuts indicate that DTBM could be a valuable tool to assess volume growth trajectories of specific white matter pathways in postnatal brain development, and potentially help in the clinical assessment of developmental delay.
Bibliography: 1) HBM, 1998, vol 6, 348-357; 2) Neuroimage 2016, 124:1125-1130; 3) Neuroimage 2016, 132:439-454;



Disclosures: **A. Nayak:** None. **M. Irfanoglu:** None. **N. Sadeghi:** None. **C. Pierpaoli:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.04/C29

Topic: A.09. Adolescent Development

Title: Age ain't nothing but a number: Salience and central executive network connectivity in high school and college students

Authors: *N. A. PADGAONKAR[1], L. E. SHERMAN[2], L. M. HERNANDEZ[1], M. DAPRETTO[1]
[1]UCLA Dept. of Psychiatry and Biobehavioral Sci., UCLA, Los Angeles, CA; [2]Dept. of Psychology, Temple Univ., Philadelphia, PA

Abstract: Functional magnetic resonance imaging (fMRI) studies show that functional connectivity in resting state networks (RSNs) develop across the lifespan. RSNs are seen as early as in utero (Schöpf et al., 2012) and develop through adolescence (Supekar et al., 2010). However, while several studies have investigated age differences in RSNs, few have considered the impact of major life transitions, such as the transition to college during late adolescence.

Also, while experience in higher education is associated with changes in structural brain connectivity (Bennett and Baird, 2006), little is known about its relationship with functional connectivity. Thus, here we focus on RSN developments across this important life transition. We examined RSNs of 28 adolescents (11 male, Mean Age = 16.9 years) in high school and 26 young adults (10 male, Mean Age = 19.8) in college. Because we were interested in both age-related effects and changes attributable to social environments (high school vs. college), we compared RSNs both with and without including age as a covariate. Based on previous work showing longitudinal changes in functional connectivity in the Default Mode Network (DMN) and Central Executive Network (CEN) during early adolescence (Sherman et al., 2014), we focused on these two networks, as well as the Salience Network (SN), which is thought to regulate the switch between DMN and CEN activity (Sridharan et al., 2008).
All participants underwent a 6-minute resting state, eyes-open scan on a 3T MRI scanner. Functional connectivity was assessed using 10 mm-diameter seed regions placed in the left precuneus, right dorsolateral prefrontal cortex (dlPFC), and right anterior insula (AI) to generate maps of the DMN, CEN, and SN respectively. No significant differences in RSNs were observed when comparing high school and college students (i.e., no effect of age). Importantly, however, when age was regressed out, significant group differences were observed in the CEN and SN, suggesting that developmental life stage was the key modulator of changes in connectivity. Specifically, college students showed greater connectivity in the CEN between the right DLPFC and the right superior parietal lobe than high school students. As compared to college students, high school students showed greater connectivity in the SN between the right AI and the right temporal pole, as well as between the right AI and the right paracingulate gyrus. These findings indicate that environmental and experiential changes taking place during the transition from late adolescence to early adulthood significantly impact the development of network connectivity above and beyond any effect of age.

Disclosures: N.A. Padgaonkar: None. **L.E. Sherman:** None. **L.M. Hernandez:** None. **M. Dapretto:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.05/C30

Topic: A.09. Adolescent Development

Support: NWO Grant PROO 411-07-153

Title: Multivariate pattern analysis shows that age-specific brain activity is shared by a broad range of cognitive tasks

Authors: *P. STIERS[1], M. BIRKISDÓTTIR[2], E. H. H. KEULERS[2]
[1]Fac. of Psychology and Neuroscience, Maastricht Univ., Maastricht, Netherlands; [2]Dept. of Neuropsychology and Psychopharmacology, Maastricht Univ., Maastricht, Netherlands

Abstract: Adolescence is characterized by dramatic changes in behavioral. Many studies have tried to pinpoint the specific neurocognitive processes that cause these behavioral changes. But, neuroanatomical studies report widespread changes in grey matter and brain connectivity that suggest a global, unspecific increase in neural efficacy that improves performance in a broad range of behavioral tasks. Consequently, age-related changes in brain activity should not be specific to particular neurocognitive processes. In line with this, we previously found that multivariate pattern classification of age from adolescents' brain activity generalizes across cognitive tasks - i.e., a classifier trained to decode age from brain activity during a go-nogo task can above chance predict age from brain activity during a complex gambling task, and vice versa (Keulers ea., 2012, NeuroImage, 60:1250-65). Despite obvious differences, these two tasks share the stimulus-response mapping structure typical of cognitive tasks. In the present study we test the generalizability of age classification between three tasks that do not share this structure: a metacognitive agenda setting task requiring a choice response based on self-reflexive evaluation; a word-pair encoding task involving a visualization strategy without an overt response; a passive visual perception task exposing participants to photos of objects, faces and movement scenes in 24 second blocks. Adolescents aged 13 (n=25) and 17 yrs. (n=22) performed the tasks in a 3T MRI scanner. For each task several condition specific percent signal change maps were computed per participant and used as examples in a support vector machine learning analysis with recursive feature elimination and leave-one-subject-out cross-validation. Within tasks, age classification accuracies were 62 to 72%, depending on task and parameters, and were significantly above chance (125 label randomization analyses). Between tasks, generalization of age decoding from the metacognitive to the encoding task was 75% accurate ($p<0.01$), while the reverse generalization yielded 69% accuracy ($p<0.01$). These accuracies were not dependent on the univariate task-responsive voxels. Generalization to the visual maps yielded 65% ($p<0.01$) when training was on metacognition and 63% ($p<0.05$) for training on encoding activity. We conclude that age-related changes in brain activation are not task specific, but common to a broad range of cognitive functions, from metacognitive judgement to passive perception. Thus, brain maturation in the teen years reflects a global increase in neural efficiency, most likely driven by increased long-range connectivity.

Disclosures: P. Stiers: None. **M. Birkisdóttir:** None. **E.H.H. Keulers:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.06/C31

Topic: A.09. Adolescent Development

Title: Development of visual P3 components from childhood to adulthood and associations with cortical structure: A study combining electrophysiology and MRI

Authors: *K. OVERBYE, R. J. HUSTER, K. B. WALHOVD, A. M. FJELL, C. K. TAMNES
Dept. of Psychology, Univ. of Oslo, Oslo, Norway

Abstract: Visual attention improves dramatically during development. The P3 component of the event related potential is assumed to indicate how the brain forms a bridge from stimulus processing to response processing, possibly related to the role of the parietal cortex in allocating attention to internal processes. However, how P3 relates to brain maturation is far from understood. In the present study, we aimed to investigate age-related differences in early visual and P3 components, and how individual differences in these components were related to cortical structure. A group of 88 healthy children and adolescents 8.2-19.7 years old completed a three-stimulus visual oddball task while high-density EEG data was recorded. Additionally, MRI data was acquired on a separate day and T1-weighted images were processed using FreeSurfer to estimate vertex-wise cortical thickness and surface area. Group-level data blind source separation was applied to EEG recordings to estimate components representative of neural sources commonly expressed across the whole sample. The resulting components included one P3 component with a parietal maximum, and also a component with a frontal maximum that in terms of latency and shape was deemed P3-like, as well as one component assumed to reflect early visual processing. Differences across age was found for all three components, with the two P3-like components being stronger for older participants while the early visual component was stronger for younger participants. Stronger P3-like components were associated with increased task speed and precision, independently of age, while the opposite was true for the strength of the early visual component. Finally, the strength of the parietal P3 was associated with a larger cortical area in the lateral temporal lobe in the left hemisphere. This was found to hold independently of age and after correction using cluster size inference. It is suggested that the age differences in the strength of components reflects the maturation of attentional mechanisms, with increased electrophysiological responses to task-relevant stimuli, representing an increased ability to effectively focus on relevant information and respond quickly and efficiently.

Disclosures: K. Overbye: None. **R.J. Huster:** None. **K.B. Walhovd:** None. **A.M. Fjell:** None. **C.K. Tamnes:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.07/C32

Topic: A.09. Adolescent Development

Support: This research was supported by the Netherlands Organisation for Scientific Research (NWO) under project number: 023.001.082

Title: Visual search becomes faster during adolescence

Authors: *R. BURGGRAAF WINGENS, ESQ[1], J. N. VAN DER GEEST[2], M. A. FRENS[3], I. HOOGE[4]
[1]Neurosci., Erasmus MC, Bosch En Duin, Netherlands; [2]Erasmus MC - Dept Neurosci. (ee12), Rotterdam, Netherlands; [3]Erasmus MC Rotterdam, Rotterdam, Netherlands; [4]applied psychology, Ignace Hooge, Utrecht, Netherlands

Abstract: A child's brain is changing tremendously during adolescence. We hypothesized that visual search will also change during this period of life. In particular, we are interested in the reaction time and accuracy of visual search and the use of selection of elements to fixate, in order to direct fixations mainly to elements most similar to the target.
A large group of adolescents (N=140; 12-19 years; 53% male) participated in a visual search experiment. Each trial showed a stimulus display with 36 Gabor patches placed on a hexagonal grid. The target consisted of a vertical oriented element with a high spatial frequency. Non-targets differed from the target in spatial frequency and/or orientation. The experiment was composed of 144 trials (50% target present) and participant had to decide whether the target was present or not.
Search performance and behavior changed during adolescence. Shorter fixation durations coincided with decreasing reaction time. The number of fixations and the selection of elements to fixate did not change, which could possibly explain the finding that search accuracy did not change with age.
In general we conclude that the process of gathering visual information does not change during adolescence, but children become faster in processing this information.

Disclosures: R. Burggraaf Wingens: None. **J.N. Van Der Geest:** None. **M.A. Frens:** None. **I. Hooge:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.08/C33

Topic: A.09. Adolescent Development

Support: NIMH R01 MH080243

Title: Context-dependent neurodevelopment of vta-accumbens connectivity: A longitudinal fmri study

Authors: *V. P. MURTY[1], D. F. MONTEZ[2], W. F. FORAN[2], F. J. CALABRO[3], B. LUNA[4]
[1]Psychiatry, [3]Departments of Psychiatry and Bioengineering, [2]Univ. of Pittsburgh, Pittsburgh, PA; [4]Dept Psychiatry, Lab. Of Neurocognitive Develop., Pittsburgh, PA

Abstract: The mesolimbic dopamine system continues to mature throughout adolescence into early adulthood. Neuroimaging has characterized increases in ventral striatal activation across adolescence during reward-motivated behavior; however, relatively less work has investigated interactions between the ventral tegmental area, the source of mesolimbic neurons, and its targets in the striatum. In the current study, we characterized this neural circuit in children, adolescence, and adults in rewarding and neutral contexts using an accelerated longitudinal fMRI design. fMRI data was collected in 170 individuals ranging in age between 10-30 years old. Participants completed both a resting-state (neutral context) and reward-motivated anti-saccade task (rewarding context) over the course of 1-3 visits. Visits were separated by approximately 1 year. To compare results across tasks, we characterized 'background connectivity' of the reward motivated anti-task, which reflects intrinsic connectivity between regions that are related to task contexts. Longitudinal fMRI analyses revealed a significant decrease in VTA-nucleus accumbens coupling in rewarding contexts as individual's approached adulthood ($p < 0.001$). Conversely, there were no differences in VTA-nucleus accumbens as a function of age in neutral contexts ($p= 0.93$; difference in contexts $p < 0.01$). These findings support a model by which connectivity of the VTA with its mesolimbic targets is relatively stable across adolescence, however, the ability to engage this circuit in motivationally relevant contexts continues to mature into early adulthood.

Disclosures: V.P. Murty: None. **D.F. Montez:** None. **W.F. Foran:** None. **F.J. Calabro:** None. **B. Luna:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.09/C34

Topic: A.09. Adolescent Development

Title: Association of sex and pubertal status on cortical thickness in typically-developing children

Authors: *J. BARONE[1], K. M. REDING[1], J. S. KIPPENHAN[1], S.-M. WEI[1], T. NASH[1], M. ZAWADZKI[1], A. BOROSHOK[1], S. MURRAY[1], H. RAAB[1], P. MARTINEZ[2], D. E. BOYLE[3], E. ROBINSON[3], P. KOHN[1], L. NIEMAN[4], P. J. SCHMIDT[2], K. F. BERMAN[1]
[2]Behavioral Endocrinol. Br., [1]Natl. Inst. of Mental Hlth., Bethesda, MD; [3]Warren Grant Magnuson Clin. Ctr., NIH, Bethesda, MD; [4]Natl. Inst. of Diabetes and Digestive and Kidney Dis., Bethesda, MD

Abstract: Background: Adolescence is a time of increased neuropsychiatric disease onset, and a critical period for the emergence of sex differences in these disorders. Neuroimaging studies of brain development document structural and functional changes during adolescence, but often assume that observed age- and sex-related differences coincide with the pubertal transition and subsequent rise in sex hormones. Here, to better understand both sex and pubertal effects, we examined cortical thickness (CT) both before and after pubertal onset as defined by clinician-rated Tanner Staging (TS). **Methods:** Eighty typically-developing children and adolescents were categorized by pubertal status: prepubertal (TS1, N=48, 8.7±0.3yrs; 18 girls) and pubertal (TS2-5, N=32, 13.0±0.7yrs; 15 girls). Multi-echo MPRAGE scans acquired on a GE 3T MRI scanner were processed with FreeSurfer v5.3. CT was computed as the distance between pial and white matter surfaces, resampled to a standardized mesh using SUMA, and smoothed with a 25mm kernel along the cortical ribbon. Data were analyzed in a 2x2 ANOVA with sex and pubertal group (prepubertal [T1] vs, pubertal [T2-5]) as between-subjects variables. For clusters showing effects of pubertal status, we ran post-hoc partial correlations between CT and individual TSs (1-5), controlling for age. For clusters showing interaction effects, we ran pairwise comparisions between pubertal groups within each sex, and between sex in each pubertal group. **Results:** We identified robust effects of pubertal status ($p<.05$, FDR-corrected) in the prefrontal cortex (PFC), sensorimotor cortex, and dorsal and ventral visual processing streams. These regions all showed greater prepubertal than pubertal CT. Post-hoc partial correlations between individual TSs and CT, controlling for age, showed a trend-level negative relationship in left frontal cortex and right dorsal stream regions ($p's=.074$). There was a main effect of sex ($p<.001$, uncorrected) with greater parietal and occipital CT in boys in both pubertal groups. Sex by pubertal status effects ($p<.001$, uncorrected) were seen in the medial(m) PFC, and both dorsal and rostral anterior cingulate cortex (dACC, rACC). Post-hoc pairwise comparisons suggest that for all these regions CT decreased with pubertal status in girls (all $p's<.001$) while no differences were seen in boys. Additionally, in the mPFC and dACC, girls showed greater CT compared to boys within the prepubertal group only. **Discussion:** Our data suggest that CT may decrease as a function of TS, controlling for age, and that prefrontal and cingulate regions show sex differences in CT both prepubertally and across puberty.

Disclosures: J. Barone: None. **K.M. Reding:** None. **J.S. Kippenhan:** None. **S. Wei:** None. **T. Nash:** None. **M. Zawadzki:** None. **A. Boroshok:** None. **S. Murray:** None. **H. Raab:** None. **P. Martinez:** None. **D.E. Boyle:** None. **E. Robinson:** None. **P. Kohn:** None. **L. Nieman:** None. **P.J. Schmidt:** None. **K.F. Berman:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.10/C35

Topic: A.09. Adolescent Development

Support: NICHD Grant F32 HD089514-01A1

NIMG Grant R01-MH103291

Brain and Behavior Foundation NARSAD Early Investigator Award

Early Career Research Fellowship from the Jacobs Foundation

IMHRO Rising Star Award

Title: Socioeconomic status and brain structure and function across development: A multimodal investigation

Authors: *M. L. ROSEN[1], *M. L. ROSEN[1], M. A. SHERIDAN[3], K. A. SAMBROOK[2], A. N. MELTZOFF[1], K. A. MCLAUGHLIN[1]
[1]Dept. of Psychology, [2]Radiology, Univ. of Washington, Seattle, WA; [3]Psychology, Univ. of North Carolina, Chapel Hill, Chapel Hill, NC

Abstract: Growing evidence suggests that childhood socioeconomic status (SES) influences neural development, which may contribute to well-documented SES-related disparities in academic achievement. Here, we apply a multimodal neuroimaging approach to investigate the associations of childhood SES with cortical structure, white matter microstructure, and neural function during a WM task across development. The sample included 66 children and adolescents (age 6-19 years). Higher SES was associated with better academic achievement and performance on the WM task. SES was unrelated to cortical thickness after FDR-correction. However, higher family income-to-need ratio was also associated with greater fractional anisotropy in the right and left superior longitudinal fasciculi (SLF). Greater connectivity in left SLF, in turn, was associated with higher academic achievement. Additionally, higher income-to-needs was associated with greater BOLD signal across many regions in the prefrontal and temporal cortex during WM encoding, maintenance, and retrieval; greater activation in these regions was associated with better task performance and higher levels of academic achievement. Taken together, these findings suggest potential neural mechanisms that link SES with both WM performance and academic achievement, including prefrontal cortex activation and the integrity of white matter connectivity within the frontoparietal network.

Disclosures: M.L. Rosen: None. **M.A. Sheridan:** None. **K.A. Sambrook:** None. **A.N. Meltzoff:** None. **K.A. McLaughlin:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.11/C36

Topic: A.09. Adolescent Development

Title: Socioeconomic status and minority status effects on brain structure and cognitive function: A multivariate analysis of the PING study dataset

Authors: *C. CARDENAS-INIGUEZ, M. G. BERMAN
Univ. of Chicago, Chicago, IL

Abstract: Childhood low socioeconomic status (SES), widely associated with increased psychosocial and environmental stress, has been previously associated with differences in brain morphometry and cognitive function. The current study investigates this finding further by using multivariate analysis approaches, such as Partial Least Squares (PLS) and Canonical Correlations, to explore the relationship between cortical thickness and cortical surface area, and conditions associated with SES, such as household income and parental education, in addition to racial/ethnic minority status, also previously associated with increased psychosocial and environmental stress. Drawing from the Pediatric Imaging, Neurocognition and Genetics dataset (PING), a dataset of 1108 diverse participants ages 3-20 (570 male, 538 female), we analyzed cortical thickness and cortical area data parcellated into lobular regions (4 per hemisphere for a total of 8 regions) using a data-driven, fuzzy clustering technique highlighting genetic similarities in cortical areas using Freesurfer. We implemented Partial Least Squares (PLS) and Canonical Correlation analyses so as to uncover significant relationships between cortical morphometry across various cortical regions and multiple demographic variables. Consistent with previous investigations in the literature, our initial PLS analyses on cortical surface area revealed that even after controlling for age and gender, all cortical lobe areas (bilateral parietal, occipital, temporal, and frontal) showed a reliable and significant positive relationship with household income and parental education, such that lower SES was associated with a smaller surface area. We did not find a significant or reliable relationship between SES measures and cortical thickness using PLS. Canonical Correlation analyses on cortical thickness and demographic variables revealed a latent variable showing a strong negative relationship between cortical thickness across all lobes and age, but a positive relationship with being White and having higher parental education. Canonical Correlation analyses on cortical surface and demographic variables revealed a latent variable showing a strong positive relationship with being White and having both higher income and higher parental education and being male, but a strong negative relationship with being Black. Our multivariate analyses are consistent with previous reports indicating childhood SES during has a differential impact on brain development, while also showing evidence to support that minority status may also have an impact on brain morphometry.

Disclosures: C. Cardenas-Iniguez: None. **M.G. Berman:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.12/C37

Topic: A.09. Adolescent Development

Support: NIH Grant MH067924

Title: Developmental stabilization of neural gain signals improves mean behavioral performance and behavioral variability

Authors: *D. F. MONTEZ[1], B. LUNA[2]
[1]Psychiatry, Univ. of Pittsburgh, Pittsburgh, PA; [2]Dept Psychiatry, Lab. Of Neurocognitive Develop., Pittsburgh, PA

Abstract: Introduction
Behavioral variability is an important barometer of cognitive functioning. During adolescent development behavioral responses both improve on average as well as stabilize. Mechanistically accounting for the stabilization of behavior is critical to our understanding of adolescent neural development. Here, we report results from a longitudinal working memory study performed over 10 years. We develop a computational model of memory-guided saccade (MGS) performance and demonstrate that improvements in mean behavioral performance and behavioral variability can be accounted for solely in terms of stabilizing neural variability.
Methods
We studied and accelerated longitudinal cohort of 126 subjects between the ages of 8 and 33 years as they performed a variant of the memory-guided saccade task. We developed a computational model of behavioral responses based on a high-dimensional drift-diffusion race model framework that incorporates variability in gain signals. The resulting model allows us to model reaction times as well as continuous metrics of response accuracy in the memory-guided saccade task
Results
We find that behavioral performance in the memory-guided saccade task improves and stabilizes during adolescence. By incorporating multiple sources of independent gain variability in a high-dimensional drift diffusion race model that we can account for the improvements in mean behavior and behavioral variability that are observed during adolescent development. Analysis of the trial-to-trial relationship between memory-guided saccade reaction times and accuracies reveals a peculiar U-shaped speed-accuracy relationship. Further analysis shows that this relationship can be accounted for by a balance of independent variability affecting working memory and response threshold gain signals.
Conclusion

Results from our computational model indicate that independent trial-to-trial variability in gain signals that affect working memory maintenance and response thresholds can account for peculiar speed-accuracy relationships observed in our data. Moreover, in addition to qualitatively accounting for the stabilization of behavioral responses, e.g. reaction time variability and saccade precision, our computational model of memory-guided saccade performance can account for improvement in mean reaction time. Thus, it appears that changes in both mean behavioral performance and behavioral variability observed during development may be the results of stabilizing of neural gain signals.

Disclosures: D.F. Montez: None. **B. Luna:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.13/C38

Topic: A.09. Adolescent Development

Support: European Union Seventh Framework ProgramFP7/2007–2013 under grant agreement nr. 266044

European Union Seventh Framework ProgramFP7/2007–2013 under grant agreement nr. 266408

European Union Seventh Framework ProgramFP7/2007–2013 under grant agreement nr. 607310

Title: Pubertal stage and body weight inversely affect activation in inhibitory areas during food choice in children

Authors: *F. VAN MEER[1], L. N. VAN DER LAAN[1], R. A. ADAN[2], G. EIBEN[3], L. LISSNER[3], M. WOLTERS[4], S. RACH[4], M. HERRMANN[5], P. ERHARD[5], D. A. MOLNAR[6], E. KOVACS[6], G. ORSI[7], P. A. M. SMEETS[1]
[1]Image Sci. Inst., [2]Translational Neuroscience, Brain Ctr. Rudolf Magnus, Univ. Med. Ctr. Utrecht, Utrecht, Netherlands; [3]Section for Epidemiology and Social Medicine, Sahlgrenska Acad., Univ. of Gothenburg, Gothenburg, Sweden; [4]Leibniz- Inst. für Präventionsforschung und Epidemiologie - BIPS, Bremen, Germany; [5]Dept. of Neuropsychology and Behavioral Neurobio., Univ. of Bremen, Bremen, Germany; [6]Dept. of Paediatrics, Univ. of Pécs, Pécs, Hungary; [7]Pécs Diagnos. Ctr., Pécs, Hungary

Abstract: Childhood obesity is a rising problem caused in part by unhealthy food choices. Food choices are based on a value signal encoded in the ventromedial prefrontal cortex (vmPFC), and self-control involves modulation of this signal by the dorsolateral prefrontal cortex (dlPFC). We

examined the effects of development, body weight and body weight history on the neural correlates of healthy food choice in children. 141 children from the I.Family/Idefics cohort (aged 10-17y) from Germany, Hungary and Sweden performed a food choice task while being scanned with fMRI and provided health and taste ratings of the foods. During the choice task participants considered either the healthiness or tastiness of the food or chose naturally. Overall, children made healthier choices when asked to consider healthiness. However, younger children and children who had gained weight chose less healthy foods when considering healthiness. dlPFC activation during food choice correlated positively with pubertal development and negatively with BMI (Fig. 1). There was a positive correlation between weight change and visual processing activation in the bilateral calcarine sulcus and lingual gyrus. When considering healthiness, cerebellum activation correlated positively with BMI and negatively with pubertal stage, and there was a positive correlation between weight change and activation in the inferior parietal gyrus. In conclusion, in younger children and children who have gained weight the positive effect of considering the healthiness of foods on their choices was smaller. Furthermore, younger children and children with a higher body weight had less activation in an area important for self-control during food choice. Pubertal development, current body weight and body weight change influenced the effect of health considerations on different areas involved in saliency. Thus the effectiveness of interventions that rely on self-control or that call attention to health aspects of food may be diminished in younger children, children who are gaining weight and overweight children.

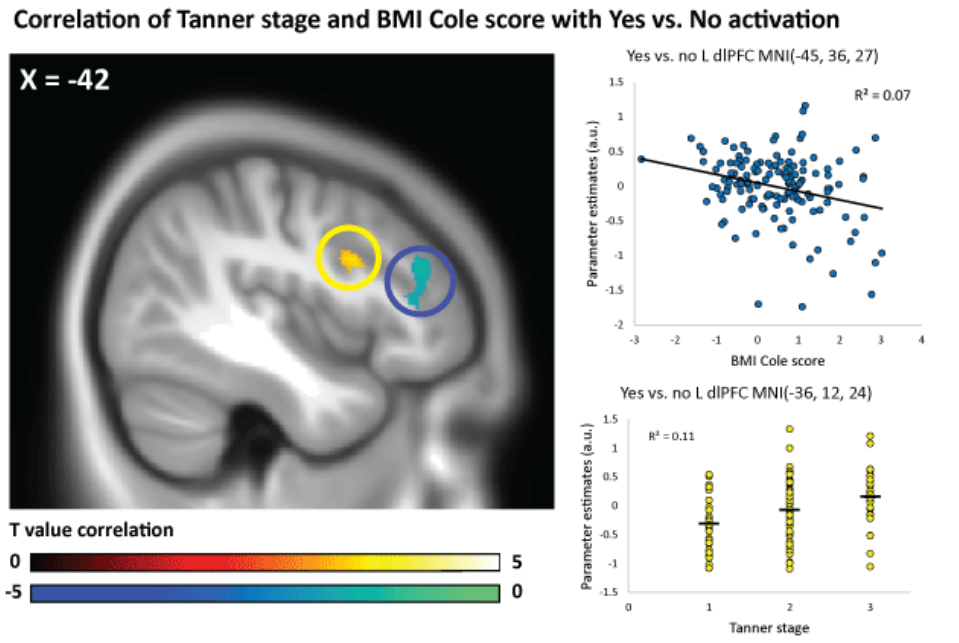


Figure 1. Correlation of Tanner stage and BMI Cole score with modulation of choice activation by yes vs. no. Blue circle denotes negative correlation with BMI Cole score and yellow circle denotes positive correlation with pubertal stage (Tanner stage). Peaks listed are significant at $p < 0.05$ level based cluster level corrections (individual voxel threshold = $p < 0.001$, cluster extent = 30 voxels, 3x3x3 mm voxels).

Disclosures: **F. Van Meer:** None. **L.N. van der Laan:** None. **R.A. Adan:** None. **G. Eiben:** None. **L. Lissner:** None. **M. Wolters:** None. **S. Rach:** None. **M. Herrmann:** None. **P. Erhard:** None. **D.A. Molnar:** None. **E. Kovacs:** None. **G. Orsi:** None. **P.A.M. Smeets:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.14/C39

Topic: A.09. Adolescent Development

Support: R01 AA017664 (Nagel)

Title: The association between cortical maturation and the development of impulsive choice in binge-drinking adolescents

Authors: *S. A. JONES[1], A. MORALES[3], B. J. NAGEL[2]
[2]Psychiatry & Behavioral Neurosci., [1]Oregon Hlth. and Sci. Univ., Portland, OR; [3]Oregon Hlth. & Sci. Univ., Portland, OR

Abstract: Previous research has shown a developmental decrease in impulsive choice across adolescence that is diminished or absent in youth who engage in binge drinking. Further, magnetic resonance imaging (MRI) has shown lateral regions of the prefrontal cortex to be associated with impulsive choice behavior. However, few studies have investigated how cortical maturation is associated with the development of impulsive choice across adolescence, and no study to our knowledge has investigated how this association differs in the context of alcohol use. In the current study, 25 binge-drinking adolescents (≥4 drinks/occasion, ≥3 times in the past 3 months) and 21 controls (matched at baseline for age, gender, and pubertal development) completed a baseline T1-weighted MRI and delay discounting procedures, prior to alcohol use, and again at follow-up (mean time between scans = 2.8 years). Multilevel modeling was used to assess the association between cortical thickness in the dorsolateral prefrontal cortex (DLPFC), and the development of delay discounting behavior across age in both binge-drinking adolescents and controls. In the right rostral middle frontal gyrus, there was a significant association between cortical thickness and discounting behavior (i.e. impulsive choice) that varied across age in control adolescents (b = -2.25, $p < 0.05$), such that less thickness earlier in adolescence (age 14) was associated with significantly lower impulsive choice (b = 7.85, $p < 0.01$), an effect that diminished and was non-significant by late adolescence/early adulthood. The association of this interaction effect, between cortical thickness and age, with impulsive choice was significantly altered in binge-drinking adolescents (group-by-thickness-by-age interaction, b = 3.39, $p < 0.05$). As a result, binge-drinking youth showed no association between cortical thickness and impulsive choice across adolescence. Together, these findings demonstrate that thinner DLPFC earlier in adolescence, representative of a developmentally typical synaptic pruning process, is associated with lower rates of impulsive choice, an effect that diminished across adolescence into early adulthood in non-alcohol users. However, cortical thickness was not associated with the development of impulsive choice in those who went on to drink, suggesting a disconnect in the

relationship between cortical thickness and impulsive choice behavior, which precedes initiation of alcohol use. This disruption in structure-function may render adolescents more prone to engage in risky decision making, such as the decision to drink.

Disclosures: S.A. Jones: None. **A. Morales:** None. **B.J. Nagel:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.15/C40

Topic: A.09. Adolescent Development

Support: CIHR 238722

Title: Early adolescent striatal and midbrain resting state connectivity correlate with impulsivity, sensation-seeking, and body mass index

Authors: *R. J. SHARKEY[1], J. BOURQUE[6], K. LARCHER[2], Y. ZHANG[2], A. ALTINKAYA[2], A. SADIKOT[3], P. CONROD[6,7], A. C. EVANS[2], H. GARAVAN[8], M. LEYTON[4], J. R. SEGUIN[6], R. PIHL[5], A. DAGHER[3]
[1]Integrated Program in Neurosci., [2]McConnell Brain Imaging Ctr., [3]Neurol. and Neurosurg., [4]Dept. of Psychiatry, [5]Dept. of Psychology, Montreal Neurolog. Inst., Montreal, QC, Canada; [6]CHU Hop. Ste-Justine, Univ. de Montreal, Montreal, QC, Canada; [7]Inst. of Psychiatry, Psychology and Neurosci., Kings Col. London, London, United Kingdom; [8]Psychiatry, Univ. of Vermont, Burlington, VT

Abstract: Across age groups, differences in connectivity between the striatum and prefrontal cortex co-vary with trait impulsivity (IMP) and sensation-seeking (SS). IMP and SS are also known to increase during early adolescence as maturation of subcortical structures outpaces that of the prefrontal cortex. While an imbalance between the striatum and prefrontal cortex is considered a normal developmental process, higher levels of adolescent IMP and SS are associated with an increased risk for diverse problems, including obesity. To examine these associations further we assessed relationships between body weight, IMP, SS, and connectivity of nuclei in the striatum and dopaminergic midbrain in young adolescents. Data were collected from 116 children between the ages of 12 and 14, and included resting state functional magnetic resonance imaging, personality measures from the Substance Use Risk Profile Scale, and body mass index Z-score for age (BMIZ). Voxel-wise analyses indicated that the connectivity of the left and right sub-thalamic nucleus (STN), ventral striatum (VS), ventral tegmental area (VTA) and substantia nigra (SN) with other cortical and limbic regions of the brain, were correlated with BMIZ, IMP and SS. The voxel-wise connectivity data was reduced by calculating the mean connectivity for each of 135 anatomical atlas regions, for each of the four seeds. The shared

variance for the connectivity of each seed, and the IMP, SS and BMIZ measures, were then analyzed using partial least squares correlations. This analysis identified a single significant striato-limbic network that was connected with the VS, VTA and SN ($p = 0.006$). This limbic network, defined using a bootstrap cut-off ratio of -3 or less, included the bilateral hippocampi, amygdalae, medial anterior temporal lobes, parahippocampal gyri and red nuclei, as well as the left perirhinal cortex, thalamus, SN and STN. Connectivity between these regions and the striato-limbic network was correlated positively with IMP and BMIZ scores and inversely with SS. Finally, results from the initial voxel-wise analysis were used to generate reverse inference maps in the NeuroSynth database. This analysis found that the voxel-based regions had been identified in prior studies on reward and risk-taking. Together, these findings emphasize that, in addition to the well-established role that striato-frontal circuits play in the development of adolescent personality traits, connectivity of limbic regions with the striatum and midbrain also impact IMP, SS and eating behaviour in adolescents.

Disclosures: R.J. Sharkey: None. **J. Bourque:** None. **K. Larcher:** None. **Y. Zhang:** None. **A. Altinkaya:** None. **A. Sadikot:** None. **P. Conrod:** None. **A.C. Evans:** None. **H. Garavan:** None. **M. Leyton:** None. **J.R. Seguin:** None. **R. Pihl:** None. **A. Dagher:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.16/C41

Topic: A.09. Adolescent Development

Support: NIH/NIAAA 1R01AA01998301

Title: Sexual dimorphism of the effect of DRD2 AUD-risk genotype on gray matter volume (GMV) and adolescent substance initiation

Authors: *R. A. SCHROEDER[1], B. W. STEVENS[1,2], V. L. DARCEY[1,2], E. J. ROSE[3], D. H. FISHBEIN[3], J. W. VANMETER[1]
[1]Ctr. For Functional and Mol. Imaging, [2]Interdisciplinary Program in Neurosci., Georgetown Univ., Washington, DC; [3]The Pennsylvania State Univ., University Park, PA

Abstract: Adolescence is a period of development marked by significant brain and behavioral changes that are further influenced by genetic and sexually dimorphic factors. One source of variation is the single nucleotide polymorphism rs6277 in the dopamine receptor D2 (DRD2) gene, in which the C allele has shown an increased risk for Alcohol Use Disorder (Padmanabhan & Luna, 2014). Since little is known regarding the effects of DRD2 genotype and sex during adolescence, we analyzed gray matter volume (GMV), future initiation of drug or alcohol use, and reward-based planning behavior.

Data are from healthy, drug and alcohol naïve adolescents, ages 11-13, N = 101 (55 F), enrolled in the Adolescent Development Study, a prospective longitudinal neuroimaging study of drug and alcohol use in adolescents. Significant differences were found between high and low risk DRD2 allele groups in race ($p < 0.001$), IQ ($p < 0.001$) and SES ($p = 0.003$), with the latter two significantly correlated ($p < 0.001$).
GMV was quantified using a T1-weighted MPRAGE (voxel size = 1mm3) acquired on a Siemens 3T scanner. GMV was assessed using voxel-based morphometry using DARTEL performed in SPM8. Statistical modeling compared groups using a 2x2 ANOVA including sex and DRD2 (T/T combined with T/C vs. C/C), while controlling for IQ and race. Intracranial brain volume was used to normalize results to avoid volume-based sex differences and multiple comparisons were controlled using Non-Stationary Cluster Correction.
No main effects for DRD2 or sex were found, however, whole brain analysis revealed an interaction between DRD2 and sex, with greater GMV found in low risk females and high risk males when compared to high risk females and low risk males (723 voxels, $p = 0.045$) in the left superior temporal gyrus (STG; peak -46, -19, -1), which has been shown to be involved with reward planning, impulsivity, and goal achievement (FitzGerald et al., 2014; Marsh et al., 2010; Gerlach et al., 2014). Within sexes, males had significant differences between high and low risk ($p < 0.001$).
Temporal discounting, a measure of reward-based planning, also had a significant interaction between sex and DRD2 genotype ($p = 0.004$), with high risk indicating a preference for immediate reward. To measure the influence of DRD2 on drug and alcohol use, an odds ratio determined that future male users with high risk versus low risk genotype were not significantly different. However, females with the low risk genotype were significantly more likely to become users (12 low risk vs. 6 high risk) at $p = 0.019$. These results suggest DRD2 has a sexually dimorphic effect on adolescent substance initiation that could be related to its differential effect on GMV in the STG.

Disclosures: R.A. Schroeder: None. **B.W. Stevens:** None. **V.L. Darcey:** None. **E.J. Rose:** None. **D.H. Fishbein:** None. **J.W. VanMeter:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.17/C42

Topic: A.09. Adolescent Development

Title: Dimensional relationships between basal ganglia iron and psychosis-spectrum symptoms in youth

Authors: *L. M. BEARD[1], A. ROSEN[2], M. A. ELLIOT[2], D. R. ROALF[3], K. PRABHAKARAN[2], R. CIRIC[4], M. E. CALKINS[2], T. M. MOORE[2], K. RUPAREL[2], R. GUR[2], R. C. GUR[5], D. H. WOLF[2], T. D. SATTERTHWAITE[2]
[1]Neuropsychiatry Section, Brain Behavior Laboratory, Hosp. of the Univers, Philadelphia, PA; [3]Neuropsychiatry, [2]Univ. of Pennsylvania, Philadelphia, PA; [4]Pomona Col., Claremont, CA; [5]Univ. Pennsylvania, Philadelphia, PA

Abstract: Elevated levels of dopamine in the basal ganglia is thought to be a critical mechanism for the development and maintenance of psychosis. Iron is implicated in dopamine transport, and individual differences in basal ganglia iron have been associated with variation in extracellular dopamine levels and reuptake. Iron can be measured non-invasively using R2* weighted magnetic resonance imaging (MRI), allowing in vivo quantification. Prior work has suggested that lower R2* signal is associated with lower iron content, which is thought to correspond to higher extracellular dopamine levels. Furthermore, it is known that basal ganglia iron and dopamine evolve dramatically during adolescence, which is also when symptoms of psychosis typically emerge. Here, we used R2* MRI to quantify basal ganglia iron in a large sample of youth; ages 8-23 imaged as part of the Philadelphia Neurodevelopmental Cohort (PNC, n=1,147, 54% female). We hypothesized that greater levels of psychosis-spectrum symptoms would be associated with lower R2*, which is consistent with higher extracellular dopamine. To test this, we used an advanced multi-atlas labeling procedure to derive subject-specific segmentations of each basal ganglia structure. Mean R2* signal in each structure was modeled using generalized additive models with penalized splines to capture both linear and nonlinear developmental effects. Replicating prior work, R2* increased with age ($p<0.001$ for all basal ganglia regions). Critically, lower R2* signal was associated with higher psychosis-spectrum symptoms in the bilateral putamen ($p<0.002$), consistent with higher extracellular dopamine. This result provides novel evidence for dopamine dysregulation associated with subthreshold psychotic symptoms during adolescence and highlights the utility of this non-invasive proxy of basal ganglia dopamine.

Disclosures: L.M. Beard: None. **A. Rosen:** None. **M.A. Elliot:** None. **D.R. Roalf:** None. **K. Prabhakaran:** None. **R. Ciric:** None. **M.E. Calkins:** None. **T.M. Moore:** None. **K. Ruparel:** None. **R. Gur:** None. **R.C. Gur:** None. **D.H. Wolf:** None. **T.D. Satterthwaite:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.18/C43

Topic: A.09. Adolescent Development

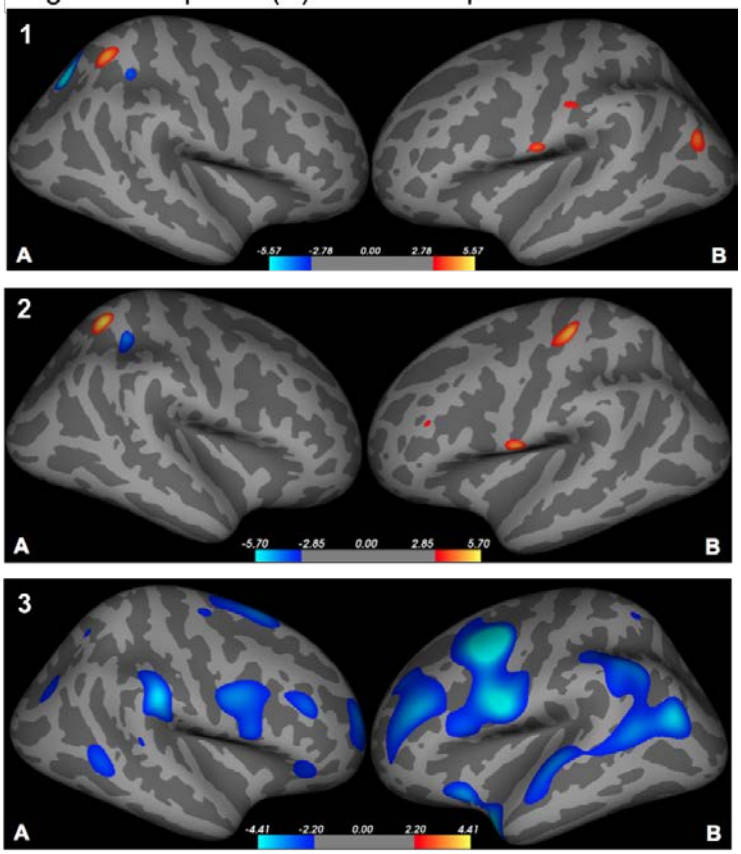
Support: 2T32MH067564

Title: Abnormal gyrification and cortical thickness in the schizophrenia prodrome: A developmental perspective

Authors: *K. DAMME[1,2], T. D. GUPTA, 60208[2], J. BERNARD[3], D. DEAN[3], R. NUSSLOCK[2], V. MITTAL[2]
[1]Northwestern Univ., Chicago, IL; [2]Psychology, Northwestern Univ., Evanston, IL; [3]Univ. of Colorado, Boulder, CO

Abstract: Psychotic disorders are characterized by abnormal cortical features, which may be informative about timing of developmental insult or specific pathogenic factors. It is unclear is if cortical abnormalities primarily manifest after onset or reflect abnormal neurodevelopment. With regard to the latter, it is unclear if they carry over from in utero development, or if in fact, emerged/changed during the significant adolescent neuro-reorganization that overlaps with the psychosis prodrome period. To date, a handful of studies in this prodromal period focus on change over time, but none have looked at the range of available surface characteristics (e.g., sulcal depth, curvature, thickness) in the same sample, over time.
A total of 43 ultra high-risk (meeting criteria for a prodromal syndrome; UHR) and 38 Healthy Control (HC) participants completed a structural scan at two-time points. Freesurfer v.5.3.0 automatic segmented structural images, T1-MPRAGE. Right and left hemispheres were tested separately and FDR $p<.05$ using QDEC to control for gender and medication status, comparing cortical thickness, curvature, and sulcal depth.
In a whole brain analyses, the UHR and HC groups differed in curvature and sulcal depth revealing nine significant clusters. UHR and HC groups differed in cortical thickness in several significant clusters as well. Group differences survived correction for age, and medication status. Sulcal depth and curvature demonstrate a similar regional pattern of gyrification distinguishing UHR from typically developing controls, consistent with a distinguishing early developmental insult. Cortical thickness represented a lager unique pattern of cortical thinning related to later adolescent neuro-reorganization. The present findings play a vital role in helping to reconcile the body of literature highlighting cortical feature anomalies in psychosis, with the neurodevelopmental perspective of schizophrenia.

Whole Brain Analyses Comparing UHR to HC FDR - corrected p<0.05: **Figure 1. Curvature Index** (A) Left Hemisphere (B) Right Hemisphere; **Figure 2. Sulcal Depth** (A) Right Hemisphere (B) Left Hemisphere **Figure 3. Cortical Thickness** (A) Right Hemisphere (B) Left Hemisphere



Disclosures: **K. Damme:** None. **T.D. Gupta:** None. **J. Bernard:** None. **D. Dean:** None. **R. Nusslock:** None. **V. Mittal:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.19/C44

Topic: A.09. Adolescent Development

Support: MQ Fellows Award to JLR

R01MH101425 to JLR

NIH Grant MH089983 to REG

NIH Grant MH089924 to REG

R01MH107703 to TDS

Title: Prenatal exposure to population-wide folic acid fortification impacts cortical maturation and psychosis risk in youth

Authors: *H. ERYILMAZ[1], F. C. HUNTINGTON[1], A. RODRIGUEZ-THOMPSON[1], T. W. SOARE[1], K. F. DOWLING[1], J. C. BLOSSOM[2], R. L. GOLLUB[1], E. SUSSER[3], R. C. GUR[4], M. E. CALKINS[4], R. E. GUR[4], T. D. SATTERTHWAITE[4], J. L. ROFFMAN[1]
[1]Psychiatry, Massachusetts Gen. Hosp., Charlestown, MA; [2]Ctr. for Geographic Analysis, Cambridge, MA; [3]Epidemiology and Psychiatry, Columbia Univ., New York, NY; [4]Univ. Pennsylvania, Philadelphia, PA

Abstract: Maturation of the cerebral cortex through adolescence supports cognitive, social, and emotional development. Disruptions of the brain's normal growth curve, such as accelerated age-related cortical thinning, characterize several neuropsychiatric conditions that emerge in the first decades of life. Epidemiologic studies of one such disorder, schizophrenia, hint that variation in gestational folate levels may influence post-natal brain development in ways salient to disease risk. To test this idea, we leveraged a "natural experiment": we contrasted cortical thickness among comparable groups of 8- to 18-year-old youth who gestated before, during, or after the rollout of mandatory grain product fortification (1996-97), a population-wide intervention that rapidly doubled blood folate levels in women of child-bearing age.

We purposely sampled the MGH Research Patient Data Registry to identify matched groups of pre-rollout, rollout, and post-rollout youth with normative clinical MRI scans that survived stringent quality control (n=315 scans). A second, deeply phenotyped cohort born around the time of the rollout (Philadelphia Neurodevelopmental Cohort, PNC, n=881 scans) was used to verify effects of fetal fortification exposure on cortical thinning, and to relate thickness changes to psychosis risk. Finally, temporal specificity of these effects was established by using a third cohort (NIH MRI Study of Brain Development, NIH) comprising 385 scans from youth born exclusively before the rollout.

Within the MGH cohort, we observed diffuse pre- to post-rollout increases in cortical thickness in 29 of 68 cortical regions-of-interest ($p<.05$, FDR corrected). The post-rollout group also exhibited delayed cortical thinning in 14 of these regions ($p<.05$, FDR corrected). This pattern replicated in 7 regions within the PNC sample ($p<.05$), with delayed thinning apparent in the post-rollout group; but was only seen in 1 region in the NIH sample. Within the PNC sample, flatter thinning profiles in each replicated region (bilateral inferior parietal lobule; right inferior temporal, middle temporal, and precentral gyri; and left lateral occipital cortex) in turn predicted reduced psychosis risk ($0.06>p>.001$).

Programming of cortical maturation in youth is sensitive to fetal folate levels, potentially reflecting alterations in the fetal epigenetic milieu. Further, that these cortical changes predicted reduced psychosis risk may portend an important and unexpected public health benefit of folic acid fortification.

Disclosures: **H. Eryilmaz:** None. **F.C. Huntington:** None. **A. Rodriguez-Thompson:** None. **T.W. Soare:** None. **K.F. Dowling:** None. **J.C. Blossom:** None. **R.L. Gollub:** None. **E. Susser:** None. **R.C. Gur:** None. **M.E. Calkins:** None. **R.E. Gur:** None. **T.D. Satterthwaite:** None. **J.L. Roffman:** F. Consulting Fees (e.g., advisory boards); Joshua L. Roffman reports grant support and consulting income from Nestle Health Sciences/Pamlab for unrelated projects. The other authors declare no competing financial interests..

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.20/C45

Topic: A.09. Adolescent Development

Title: Brain maturation effect on functional network graph metrics of patients with attention deficit hyperactivity disorder differs from normal children and adolescents. Assortativity as a measure of brain maturation

Authors: *B. MOHAJER[1], N. ABBASI[2], A. ABDOLALIZADEH[2]
[1]Student Scientific Res. Ctr. of Tehran Unive, Tehran, Iran, Islamic Republic of; [2]Tehran Univ. of Med. Sci., Tehran, Iran, Islamic Republic of

Abstract: Introduction: Attention deficit hyperactivity disorder (ADHD), as one of the most common childhood and adolescent's neuropsychiatric disorders, has shown to demonstrate impairments of structural and functional neural connectivity networks, a finding that occurs in the maturation and pruning period of the brain synapses. Comparing the effect of aging on brain network development between healthy children and adolescents and ADHD patients would help to clarify this process.**Methods:** We used restingstate functional correlation matrices of 190 ADHD patients, and 330 age and sex matched healthy children and adolescents from ADHD200 study (umcd.humanconnectomeproject.org). Minimum, maximum and average ages were 7.17, 21.74 and 11.83 years respectively. Each matrix consisted of undirected weighted correlations between 200 regions in fMRI of subjects (Craddock atlas). Graph theoretical analyses were performed using the Brain Connectivity Toolbox. To enable comparison of global network properties across groups, we applied 20% sparsity threshold based on previous studies. Global efficiency, Transitivity, Assortativity, and Modularity were calculated as global network measures. Using R for statistical analyses, measures were fed into General Linear Model (GLM), to investigate the age effect controlled for gender effect.**Results:** We found a significant decrease in Assortativity with the increase of age in healthy subjects after FDR correction (P value=0.018, tvalue=2.3) compared to non-significant result with a slower slop of assortativity decrease in ADHD (P value=0.19, t value=1.3) patients. The remaining measures did not reveal any significant changes with the increase of age.**Discussion:** Number of neighboring nodes that

sharing an edge with a node, defines as the degree. Assortativity indicates how preferentially nodes of similar degree connect to each other. As previously shown, adult and child functional brain networks both demonstrate smallworld features, although consisting of different communities of nodes. The difference here is nodes arrangement: children's communities arranged by anatomical proximity on the other hands, adults' predominantly reflect functional relationships. Above results may indicate architectural changes in the functional network of normal population, from anatomical proximity connectivity through functional relationships. This connectivity pattern shift may be the main reason behind neural network malfunctions in ADHD, what reflects of assortativity. Future functional connectivity studies focusing on the regional measures may clarify network dynamics in ADHD population

Disclosures: B. Mohajer: None. **N. Abbasi:** None. **A. Abdolalizadeh:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.21/C46

Topic: A.09. Adolescent Development

Support: 1-ZIA-MH002860-08

Title: Amygdala response to distress cues and callous-unemotional traits; moderation by trauma

Authors: *H. MEFFERT, P. M. TYLER, M. L. BOTKIN, A. K. ERWAY, V. KOLLI, L. C. THORNTON, K. POPE, S. F. WHITE, J. R. BLAIR
Ctr. for Neurobehavioral Res., Boys Town Natl. Res. Hosp., Boys Town, NE

Abstract: Youth displaying disruptive behavior show amygdala hypo-responsiveness to fearful expressions as a function of a callous-unemotional (CU) traits (i.e., reduced guilt and empathy). However, some research suggests that trauma exposure may moderate this relationship. Specifically, work has identified two groups of disruptive youth with equivalent high levels of CU-traits, but differing levels of anxiety and trauma exposure. The objective of the first of two studies was to examine whether trauma exposure influenced the neurobiology underlying fear expression processing in 72 youth with varying levels of disruptive behavior and trauma exposure. Participants performed a gender discrimination task while viewing morphed expressions (0%, 50%, 100%, 150% fear). A linear regression analysis on the BOLD data, using level of CU-traits and trauma exposure as covariates, showed a significant CU-traits-by-trauma interaction within right amygdala; CU-traits were *negatively* associated with fear intensity modulated amygdala responses in low trauma participants, but *positively* associated with fear intensity modulated amygdala responses in high trauma participants. The second study aimed at examining how the neurobiology underpinning fear expression processing predicted social

behavior as a function of trauma exposure. Participants were invited back to complete a social goals task. Data suggest that stronger fear responsivity in the amygdala predicts prosocial behavior in low trauma youth, whereas stronger fear responsivity predicts non-social behavior (revenge) in high trauma youth. These data suggest that there is not a unique pathophysiology associated with increased CU-traits. In youth with low trauma exposure, CU-traits relate to a reduced empathic response to distress cues. This is expressed at the neural level as a reduced amygdala response to these distress cues. However, in youth with high trauma exposure, an environmental insult known to increase threat sensitivity, CU-traits relate to a heightened response to fearful expression stimuli. Importantly, the current data strongly reinforce the importance of determining the pathophysiology underpinning a particular behavioral presentation; interventions need to be tailored to the individual's pathophysiology rather than their behavioral presentation alone.

Disclosures: H. Meffert: None. **P.M. Tyler:** None. **M.L. Botkin:** None. **A.K. Erway:** None. **V. Kolli:** None. **L.C. Thornton:** None. **K. Pope:** None. **S.F. White:** None. **J.R. Blair:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.22/C47

Topic: A.09. Adolescent Development

Support: Fordham Grant 2017

Title: Age-related changes in amygdala functional connectivity in children

Authors: *T. IARAJULI, A. ROY
Neurosci., Fordham Univ., Bronx, NY

Abstract: The amygdala is a small brain region that is responsible for emotional expression, particularly fear, rage and aggression. The affective and cognitive processes involved in emotion regulation are thought to emerge from connections between the amygdala and the prefrontal cortex. Recent evidence suggests that maturation of amygdala-prefrontal circuits during childhood underlie the development of emotion regulation skills. Many studies have been conducted that compare amygdala connectivity between children and adult groups, but few have focused solely on amygdala development during middle childhood (ages 5-9 years), a time during which emotional and behavioral dysregulation often emerges. The present study used resting-state functional magnetic resonance imaging to examine the relationship between amygdala functional connectivity and age in a sample of typically developing children, ranging from 5-9 years old (n=39). We further examined how this connectivity was directly related to

emotion lability and emotion regulation using a parent-report scale, the Emotion Regulation Checklist (ERC). Results show decreases in intrinsic functional connectivity (iFC) between the right and left amygdala with increasing age. No age-related changes were observed in amygdala –PFC iFC. Additional analyses revealed that right amygdala iFC to left amygdala correlated with the ERC Lability/Negativity scale (r=0.32, p=0.04), but not ERC Emotion Regulation scale (r=-0.13, p=0.44) in typically developing children. These findings suggest that bilateral iFC of the amygdala may play a role in decreasing emotional lability across this age range.

Disclosures: T. Iarajuli: None. **A. Roy:** None.

Poster

371. Imaging of Human Brain Maturation and Mental Health

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 371.23/C48

Topic: A.09. Adolescent Development

Title: Regional developmental patterns within components of grey-/white matter intensity contrast and modulatory effects of mental health in youth

Authors: *L. C. NORBOM[1], N. T. DOAN[2], D. ALNÆS[2], T. KAUFMANN[2], L. T. WESTLYE[2], C. K. TAMNES[3]
[1]Dept. of Psychology, Univ. of Oslo, [2]NORMENT, Oslo, Norway; [3]Univ. of Oslo, 0317 Oslo, Norway

Abstract: Adolescence is a maturational period with extensive brain reorganization and coincides with the onset period of numerous mental disorders. Cortical dysmyelinisation is a possible mechanism for several of these disorders including psychotic disorders. Cholesterol in myelin is a major determinant of the signal intensity in T1-weighted MRI and the *contrast* between grey matter (GM) and white matter (WM) intensity is thus believed to be sensitive to myelin content both in WM and within the cerebral cortex. In the current study, we investigated GM/WM contrast to indirectly explore patterns of typical and atypical cortical myelinisation in adolescence, using the Philadelphia Neurodevelopmental Cohort (N=935, 8-22 years old). T1-weighted images were processed in FreeSurfer. Average GM intensities (0-60% into the cortical ribbon) and average WM intensities (0-1.5mm into subcortical WM) were sampled vertex-wise along the cortical surface, and subsequently a GM/WM contrast map was calculated. Next, we performed independent component analysis (ICA) on the contrast map, and tested for associations with age, gender, IQ, as well as a set of 7 clinical ICA components reflecting different clinical domains using linear models. Preliminary results of 10 contrast components reflected global patterns of reduced contrast with age. Over and above the global components, which explained a large portion of the data variance, regional contrast variations were reflected in a set of components, which showed either reduced or increased contrast with age. Mental

health and IQ was moderately associated with component loadings. For instance, high loading on certain clinical components were associated with increased contrast in several regions, while decreased contrast was associated with higher IQ score. Preliminary results also indicated some interaction effects between clinical components and age on GM/WM contrast. High loadings on several clinical components were associated with increased contrast for younger individuals, but not for older individuals. Our preliminary results suggest that independent modes of GM/WM contrast variation may be sensitive to global and regional maturational processes, likely related to cortical myelinisation, and that these age-related patterns to some extent are modulated by mental health and IQ.

Disclosures: L.C. Norbom: None. **N.T. Doan:** None. **D. Alnæs:** None. **T. Kaufmann:** None. **L.T. Westlye:** None. **C.K. Tamnes:** None.

Poster

372. Insights Into Developmental Vulnerabilities

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 372.01/C49

Topic: A.09. Adolescent Development

Support: NIH Grant DA039865

Hillman Foundation Seed Grant from University of Pittsburgh Brain Institute

Title: Circadian perturbation reveals time sensitivity to reward-related behavior during adolescence

Authors: *M. A. HILDEBRAND[1], C. A. VADNIE[2], R. W. LOGAN[3], M. M. TORREGROSSA[1], C. A. MCCLUNG[4]
[1]Psychiatry, [2]Dept. of Psychiatry, Univ. of Pittsburgh, Pittsburgh, PA; [3]PSYCHIATRY, UNIVERSITY OF PITTSBURGH, PITTSBURGH, PA; [4]Psychiatry, Univ. of Pittsburgh Med. Sch., Pittsburgh, PA

Abstract: Adolescence is a vulnerable time for perturbations in sleep patterns and circadian rhythms. Adolescents prefer later sleep and wake times, which has been attributed to a biologically driven delay in circadian rhythms. Since school start times are early in the morning, many adolescents may experience chronic circadian misalignment and sleep deprivation. Numerous studies support that sleep and circadian rhythm disruptions are risk factors for increased reward seeking behavior, suggesting that adolescents with poor sleep quality and misaligned rhythms may be more susceptible to uncontrolled reward seeking. Thus, we sought to determine the effects of circadian disruption on reward-related behavior in adolescent rats. The circadian rhythms and sleep patterns of adolescent male Wistar rats were monitored in non-

invasive Piezo Sleep boxes (Signal Solutions LLC) from age P25-P35 in constant dark to determine their sleep/circadian phenotype. Rats were subsequently tested for operant self-administration of sucrose pellets during their active phase from approximately age P39-P56 to determine if their motivation for reward was associated with their sleep/circadian phenotype. On the final two days, rats responded on a random ratio (RR3) schedule during their inactive phase to determine if there were associations between reward-related behavior and sleep/circadian phenotype during an acute disruption of sleep and circadian rhythms. The change in performing operant sucrose self-administration during their active phase relative to their inactive phase produced two distinct phenotypes. Time sensitive rats (n=10) decreased more than 20% in their performance, whereas time insensitive rats (n=10) had less than a 10% decrease in performance during the inactive phase. These two groups differed in their responding for sucrose reward ($p < 0.001$). The time insensitive group showed a trend for more fragmented sleep than the time sensitive group ($p = 0.093$). The time insensitive group also took longer to learn a reversal of the active lever ($p < 0.01$). Adolescent rats that had more fragmented sleep showed increased reward-related behavior during their inactive phase. This suggests that poor sleep quality might increase reward-seeking behavior. Developing a model system to understand the mechanisms underlying the associations between sleep and circadian phenotypes and reward-related behavior could allow us to develop novel treatments to reduce substance use disorders in adolescents.

Disclosures: M.A. Hildebrand: None. **C.A. Vadnie:** None. **R.W. Logan:** None. **M.M. Torregrossa:** None. **C.A. McClung:** None.

Poster

372. Insights Into Developmental Vulnerabilities

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 372.02/C50

Topic: A.09. Adolescent Development

Support: Intramural Funding (University of Massachusetts Boston)

Title: Investigating the role of CXCR-4 in the adolescent sensitization to amphetamine

Authors: *B. M. MASON, S. DONALDSON, C. CALHOUN, V. WOYTOWICZ, L. PINA, R. KANDA
Univ. of Massachusetts Boston, Boston, MA

Abstract: Drug addiction remains a significant health problem in the U.S., with about 21 million people presenting with substance use disorders. For many, the first experience with an illicit substance is in adolescence, which warrants investigative studies of drug-related behavior at this age. Neuroinflammation has recently been found to contribute to the pathology of addiction in animals, and has been implicated in the plasticity contributing to psychostimulant sensitization,

through the activation of a chemokine receptor, CXCR-4. In our current study, we investigated whether 4-day amphetamine (4.0 mg/kg/ml, IP, every 48 h) locomotor sensitization could be attenuated by the CXCR-4 antagonist, AMD3100 (5.0 mg/kg per ml) in Long-Evans adolescent males (N=39). Indeed, we observed a significant diminution in locomotor, rearing and stereotypy counts in rats pre-treated with AMD3100 prior to amphetamine administration. Further, when challenged with a low dose of amphetamine (1.0 mg/kg/ml, IP) one week later, AMD3100 pre-treatment reduced locomotor and rearing counts, and stereotypies. Additionally, we found reduced CXCR-4 counts in the medial prefrontal cortex (mPFC) of AMD3100 -pretreated rats as compared to rats given amphetamine alone. Collectively, the data suggest that amphetamine sensitization in adolescence may involve, at least in part, activation of the neuroinflammatory CXCR-4 receptor, and these lasting changes may produce the vulnerability to persistent drug-taking in adulthood.

Disclosures: B.M. Mason: None. **S. Donaldson:** None. **C. Calhoun:** None. **V. Woytowicz:** None. **L. Pina:** None. **R. Kanda:** None.

Poster

372. Insights Into Developmental Vulnerabilities

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 372.03/C51

Topic: A.09. Adolescent Development

Title: IL-1b promotes investment in present versus future outcomes

Authors: *J. GASSEN
Psychology, Texas Christian Univ., Fort Worth, TX

Abstract: Exposure to adversity early in life has been associated with risk-taking, impulsivity, and other Life history theory predicts that exposure to adversity early in life leads to the development of a faster life history strategy, characterized by investment in reproductive effort and a preference for current over future rewards. As a strong link exists between childhood adversity and measures of inflammation, we predicted that proinflammatory cytokines, such as interleukin-1 beta (IL-1β) may mediate the relationship between childhood adversity and the suite of behaviors and attitudes that together define an individual's life history strategy. We found that levels of serum IL-1β predicted more reported impulsivity, less preference for delaying gratification, and a more present temporal orientation. Analysis of *in vitro* IL-1b release by participant peripheral blood-derived mononuclear cells (PBMCs) after stimulation revealed that higher levels of IL-1b predicted a reduced expectation of future longevity. These results were not found with IL-6 or TNF-a. Taken together, these results provide evidence to suggest that proinflammatory cytokines, in particular IL-1b, are involved in shaping an individual's life history strategy.

Disclosures: J. Gassen: None.

Poster

372. Insights Into Developmental Vulnerabilities

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 372.04/C52

Topic: A.09. Adolescent Development

Support: (NIDA) R01 (NIH) DA037911

Title: Dopamine development to the orbital prefrontal cortex is protracted and sensitive to amphetamine in adolescence

Authors: *D. HOOPS[1], L. REYNOLDS[1], J.-M. RESTREPO-LOZANO[1], C. FLORES[2]
[1]Dept. of Psychiatry, [2]Dept of Psych, McGill Univ., Verdun, QC, Canada

Abstract: Introduction Exposure to drugs of abuse in adolescence alters prefrontal cortex development and adult function. We now know that exposure to abused doses of amphetamine in adolescence alters the development of the dopamine input to the medial prefrontal cortex (mPFC) and that this is dependent on the guidance cue receptor DCC. In contrast, the impact of drug exposure on orbital prefrontal cortex (oPFC) dopamine development is unknown. We do know that the maturation of oPFC-dependent behaviours depends on DCC signalling in dopamine neurons and is altered by amphetamine in adolescence. To examine the potential impact of drugs of abuse in the oPFC, we first determined whether dopamine development in the oPFC is protracted across adolescence. Then, we examined whether exposure to abused doses of amphetamine during adolescence disrupts oPFC dopamine development, and if DCC is implicated in this disruption. **Methods** We counted dopamine varicosities in the oPFCs of adolescent and adult mice using neuroanatomical methods established in our laboratory. We also counted dopamine varicosities in the adjacent piriform cortex as a control. We then treated adolescent mice with saline or amphetamine using a treatment regimen known to disrupt mPFC dopamine development via DCC. Once the mice reached adulthood, we counted dopamine varicosities and dopamine/DCC varicosities. **Results** Dopamine varicosity density doubles between early adolescence and adulthood in the lateral and insular oPFC, indicating protracted dopamine development across adolescence. In contrast, dopamine development in the piriform cortex was not protracted. Remarkably, chronic administration of amphetamine during adolescence dramatically reduces (by ~30%) adult dopamine varicosity density in these regions. This drug effect results in more DCC-expressing axons in the oPFC. **Discussion** These findings provide a potential mechanism underlying the changes in oPFC-mediated behaviours linked to adolescent drug abuse. Building on these findings, we are now using viral vector mediated cell type-specific fluorescent markers to confirm that dopamine axons are growing to the oPFC during adolescence, and where these axons are located prior to the onset of adolescence.

Disclosures: **D. Hoops:** None. **L. Reynolds:** None. **J. Restrepo-Lozano:** None. **C. Flores:** None.

Poster

372. Insights Into Developmental Vulnerabilities

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 372.05/C53

Topic: A.09. Adolescent Development

Support: NIH Grant HD077623

NIH Grant MH078105-01S1

NIH Grant MH078105-04S1

NIH Grant MH091645

NIH Grant MH086633

NIH Grant U54 HD079124

NIH Grant MH100029

Title: Social stress and the dietary environment affect infant brain volumes in macaques: Modulation by maternal factors

Authors: *M. H. KYLE[1], ***M. H. KYLE**[1], *M. H. KYLE[2], M. PINCUS[1,2], J. GODFREY[1,2], Y. SHI[3], M. STYNER[3], L. LI[1], B. R. HOWELL[4], K. ETHUN[2], M. E. WILSON[1,2], M. SANCHEZ[1,2,5]
[1]Emory Univ., Atlanta, GA; [2]Yerkes Natl. Primate Res. Ctr., Atlanta, GA; [3]Dept of Psychiatry, Univ. of North Carolina, Chapel Hill, NC; [4]Univ. of Minnesota, Inst. of Child Develop., Minneapolis, MN; [5]Ctr. for Behavioral Neurosci., Atlanta, GA

Abstract: Social stress has adverse effects on brain structure and function linked to psychopathology, including changes to amygdala and prefrontal volumes. It is not well understood how these alterations emerge during development and interact with other risk factors, including obesogenic diets. Here we examined the potential for a synergistic impact of social stress and diet on infant brain development longitudinally, using a translational macaque model of social subordination stress. We also probed whether maternal care and breast milk mediate the effects of stress and diet on neural development. Forty-four socially-housed rhesus monkey infant-dam pairs (n=21 dominant (DOM), n=23 subordinate (SUB)) were randomly assigned to either a chow-only, low-calorie diet (LCD) condition, or had access to both LCD and high-calorie diet (HCD) from birth (choice). Behavioral observations of the infant-dam pairs were

collected from birth through 6 months, and maternal care quality was also rated. Plasma cortisol and structural MRI scans were collected at 2 weeks and 6 months. Preliminary results of the 6-month imaging data reveal significantly larger overall brain volumes in infants on the choice diet, with regional increases in the caudate, and insular, cingulate, and parietal cortices. SUB infants had larger amygdala and hippocampal volumes. The SUB and the choice conditions were associated with larger white matter (WM) volume in the frontal lobe, and a diet by social rank interaction was detected for WM in temporal visual and auditory cortices, with DOM LCD infants showing the smallest volumes. Baseline cortisol at 6 months was higher in the choice than LCD condition, which predicted larger right amygdala volumes. Among several rank (and diet) effects on maternal care, those on maternal permissiveness/attachment at 2 weeks were interesting, as stronger attachment predicted smaller left amygdala volume at 6 months. Greater milk yield at 6 weeks also predicted larger brain development, particularly of amygdala, and cingulate and frontal cortices WM. These preliminary findings suggest that exposure to social subordination and obesogenic diet early in life impacts infant brain development and stress hormones. Maternal care was also influenced by rank and diet, with more competent maternal care seeming to protect against some of the negative impacts of stress and diet on brain development, specifically in the amygdala.

Disclosures: **M.H. Kyle:** None. **M. Pincus:** None. **J. Godfrey:** None. **Y. Shi:** None. **M. Styner:** None. **L. Li:** None. **B.R. Howell:** None. **K. Ethun:** None. **M.E. Wilson:** None. **M. Sanchez:** None.

Poster

372. Insights Into Developmental Vulnerabilities

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 372.06/C54

Topic: A.09. Adolescent Development

Support: NIH Grant 5R01MH104261-04

Pritzker Neuropsychiatric Research Consortium

Hope for Depression Research Foundation

ONR Grant N00014-12-1-0366

Title: Environmental Enrichment alters the behavioral phenotype of a unique animal model of mood disorders

Authors: *A. M. O'CONNOR, C. A. TURNER, E. L. AURBACH, E. K. HEBDA-BAUER, S. J. WATSON, H. AKIL
MBNI Univ. of Michigan, Ann Arbor, MI

Abstract: Selectively bred High-Responder (bHR) and bred Low-Responder (bLR) rats present a novel rodent model of mood disorders. bHR animals emulate externalizing mood disorders such as addiction and impulsivity, and bLR animals emulate internalizing mood disorders such as depression and anxiety. Environmental enrichment (EE) has previously been shown to decrease anxiety-like behavior of adult bLRs and alter social behaviors of adult bHRs. Adolescence is a critical window for the development of emotional centers within the brain, and is a time of increased negative social interactions; by early adolescence, bHR and bLR animals show differential behavioral phenotypes. The effect of EE during adolescence on emotional behavior in these lines has not yet been determined. This study investigates the impact of adolescent EE on anxiety-like behavior of bHR and bLR animals following a social stressor. Male animals from generations F49 and F53 of the colony maintained at the University of Michigan were placed into enriched environments (E) for one hour a day, 5 days a week starting at 35 days postnatal (P35) and ending at P60. Control animals were handled and placed into large cages for the same time period (C) or left in standard housing with no handling (S). Half of all animals underwent social defeat (SD) from P60 until P64. All animals underwent behavioral testing P65 - P67. P65 and P66 testing consisted of a two day social interaction assessment; on day one animals were exposed to an empty open field and on day two a novel animal was present within the open field. Elevated Plus Maze (EPM) testing was carried out on day P67. All animals were sacrificed on P68, brains collected and snap frozen for processing. Exposure to EE decreased anxiety-like behaviors in bHRs, with animals exposed to EE spending more time in the center of the open field (P=0.024) and on the open arms of the EPM (P=0.011). EE prior to SD decreased social anxiety in bLRs, with EE + SD animals showing decreased social avoidance compared to bLR S + SD animals (P=0.028). EE decreased social interaction anxiety in bHRs, regardless of SD status (P=0.020). bLR animals that experienced EE prior to SD showed “bHR-like” patterns of behavior during social interaction testing, with a similar ratio of social interaction/avoidance when a novel animal was present in the open-field. It appears that exposure to EE during adolescence “flips” the bLR phenotype regarding social behavior when animals are subject to SD, suggesting that both genotype and environment influence animals’ response to stressors.

Disclosures: **A.M. O'Connor:** None. **C.A. Turner:** None. **E.L. Aurbach:** None. **E.K. Hebda-Bauer:** None. **S.J. Watson:** None. **H. Akil:** None.

Poster

372. Insights Into Developmental Vulnerabilities

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 372.07/C55

Topic: A.09. Adolescent Development

Support: R01MH107556

Title: Transgenerational effects of early life stress: DNA methylation in brain and blood

Authors: *E. COLEY[1], P. GANGULY[1], M. S. TRIVEDI[3], H. C. BRENHOUSE[2]
[2]Psychology, [1]Northeastern Univ., Boston, MA; [3]Neuro-Pharmacology, Nova Southeastern Univ., Fort Lauderdale, FL

Abstract: Childhood adversity can compromise neural structure and function, thereby increasing susceptibility to various psychopathologies including schizophrenia and major depression. It has been shown that the hypothalamus-pituitary-adrenal (HPA) axis is dysregulated in individuals exposed to psychosocial stress at critical developmental periods. The NR3C1 gene, which encodes for the glucocorticoid receptor, is a key component in the normative functioning of the HPA axis via negative feedback on the stress response. Prior evidence from animal studies involving early life stress paradigms indicates epigenetic modulation of the NR3C1 gene. It is unclear the extent to which such epigenetic modifications are inherited by subsequent generations, and if the environment after birth compounds or ameliorates these changes. This study explores (1) the effects of early life stress (ELS) on epigenetic modification of the NR3C1 gene, and subsequent impacts on behavior and corticosterone production, (2) transmission of these modifications to following progeny, and (3) influence of early life parenting environments on such epigenetic inheritance. We used a rodent maternal separation model, which is an ethologically relevant ELS paradigm. F0 litters were either exposed to maternal separation or left undisturbed. At young adulthood (P60) rats from each group were either mated within experimental group, or subjected to behavioral paradigms including open field and social preference, to test anxiety and social behavior, respectively. Following behavioral tests, trunk blood (serum) and brain tissue (prefrontal cortex and dorsal and ventral hippocampus) were collected. F1 pups were either cross-fostered across experimental group (ELS → CON or CON → ELS), or raised by their biological mothers. The pups were allowed to grow undisturbed, and at P60, the same behavioral assessments were performed, followed by blood and brain tissue collection. Data collection strategies included: bisulfite sequencing of dorsal hippocampus tissue and serum, ELISA assay on serum to measure corticosterone concentration, RT-PCR assay on selected brain regions and serum to quantify expression of glucocorticoid receptor mRNA, and behavioral tests to assess anxiety and antisocial behaviors.Results will illuminate the transgenerational epigenetic inheritance of early life stress effects and related behaviors. Clarifying these mechanisms may inform efforts towards prevention and treatment of relevant physical and psychiatric conditions.

Disclosures: E. Coley: None. **P. Ganguly:** None. **M.S. Trivedi:** None. **H.C. Brenhouse:** None.

Poster

372. Insights Into Developmental Vulnerabilities

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 372.08/C56

Topic: A.09. Adolescent Development

Support: F32 DA043308

R01 DA034185

R01 MH101183

Title: The developmental relationship between microglia and dopamine d1 receptors in the nucleus accumbens is altered by adolescent morphine exposure

Authors: *A. M. KOPEC[1], N. R. AYRE[2], S. C. SWEAT[2], S. D. BILBO[1]
[1]Pediatrics, Harvard Med. School/Massachusetts Gen. Hospi, Charlestown, MA; [2]Duke Univ., Durham, NC

Abstract: Drug taking is often initiated in adolescence when the dopaminergic "reward circuitry," including the nucleus accumbens (NAc), is developing, and thus vulnerable to external influences. Indeed, male rats given a 5-day morphine treatment during adolescence, but not young adulthood, have persistently impaired molecular signaling in the NAc and increased risk of reinstatement (i.e. relapse) much later in life. However, how the reward circuitry develops naturally, and how addictive substances modify this development, is unknown. The immune cells of the brain, microglia, are important for normal neural development, notably via engulfment and elimination of synapses 'tagged' by the complement system of proteins, including complement protein C3 (i.e. synaptic pruning). Moreover, microglia have been implicated in playing a critical role in the adverse effects of drugs of abuse. Our data suggest that NAc dopamine D1r receptors are 'tagged' by C3 for microglial-dependent synaptic elimination during adolescent development in male rats. Five days of morphine during the adolescent period increases microglial engulfment of D1rs and reduces D1r levels after drug cessation, raising the possibility that the developmental pruning process may be over-activated by drug use. If so, increased (i) C3 levels, (ii) C3-tagged D1rs, and (iii), microglial engulfment of C3-tagged D1rs, may occur prior to D1r elimination. C3 levels are, indeed, increased after drug cessation, but there is no change in C3-tagged D1rs and decreased engulfment of C3-tagged D1rs. These data collectively suggest that adolescent morphine exposure dysregulates the C3 tagging process, thus resulting in uncoordinated and exaggerated microglial elimination of D1rs in the NAc.

Disclosures: **A.M. Kopec:** None. **N.R. Ayre:** None. **S.C. Sweat:** None. **S.D. Bilbo:** None.

Poster

372. Insights Into Developmental Vulnerabilities

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 372.09/C57

Topic: A.09. Adolescent Development

Support: NIH Grant RO1AA021517

Title: Parental alcohol exposure in binge pattern during puberty in rodent model impairs offspring development through puberty

Authors: *A. ASIMES[1], A. CUARENTA[2], C. K. KIM[1], A. P. AUGER[3], T. R. PAK[1]
[1]Cell and Mol. Physiol., Loyola Univ. Chicago Dept. of Cell and Mol. Physiol., Maywood, IL; [2]Univ. of Wisconsin, Madison, WI; [3]Psychology, Univ. Wisconsin, Madison, Madison, WI

Abstract: Parental inheritance extends beyond mere genetics, or DNA sequence, and can impact offspring traits through epigenetic mechanisms. Recent work has shown that preconception behaviors of both mothers and fathers, such as drug exposure, can impact offspring outcomes. The most widely abused drug in the United States is alcohol with more than 5.3 million Americans under the age of 21 engaging in binge pattern drinking. This type of rapid consumption during pubertal development can have long lasting effects in the brain. Recently, our lab has shown that alcohol-naïve offspring of animals exposed to alcohol during adolescence show altered DNA methylation patterns in the hypothalamus, a region of the brain involved in regulation of pubertal development, stress regulation, and behavior. Therefore, our current work aims to characterize functional consequences in these offspring. Our hypothesis is that offspring of binge-alcohol exposed parents will have altered development associated with hypothalamic function, including improper pubertal development, impaired socialization, and dysregulated stress response. We used an established rodent model of repeated adolescent binge alcohol (EtOH) exposure. Wistar rats received 3g/kg of 20% (v/v) EtOH via oral gavage once daily for 3 days, then 2 days vehicle, and another 3 days EtOH at both early and late puberty (PND37, 67). Animals were paired (EtOH-EtOH, vehicle-vehicle) for mating 24h after the last EtOH dose. Litters were culled to 10 pups per dam and were left with their biological mother until weaning at PND23 when they were separated into same-sex groupings of 5. Behavioral observations were made from recordings of home cage activity daily from PND25 - 30. Animals were then separated into paired-housing and randomly assigned to EtOH or vehicle treatment using the same adolescent binge-alcohol paradigm described above. All animals were euthanized 1h following last gavage at PND44. Cortisol levels were not significantly impacted by parental treatment, nor was expression of stress-mediator genes. However, offspring whose parents received alcohol displayed less social interaction play behaviors. Male offspring were also smaller than control counterparts, although there was no difference in weight at birth or PND7.

Subsequent alcohol exposure decreased male weights further, as well as lowered circulating testosterone and luteinizing hormone. Overall, these results show that preconception alcohol consumption by parents can impact the social and pubertal development of offspring, and can be exacerbated by offspring alcohol consumption.

Disclosures: A. Asimes: None. **A. Cuarenta:** None. **C.K. Kim:** None. **A.P. Auger:** None. **T.R. Pak:** None.

Poster

372. Insights Into Developmental Vulnerabilities

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 372.10/C58

Topic: A.09. Adolescent Development

Support: NIH Grant MH099625

NIH Training Grant T32 ES007326

Title: Stress during adolescence does not alter the total number or morphology of microglia in the male or female mPFC in adulthood

Authors: *C. DRZEWIECKI[1], J. WILLING[2], L. CORTES[3], J. M. JURASKA[2]
[1]Univ. of Illinois at Urbana/Champaign, Champaign, IL; [2]Psychology, [3]Univ. of Illinois, Champaign, IL

Abstract: Adolescents are particularly susceptible to the effects of stress, though the neurobiological basis of this vulnerability remains unknown. Stress in adolescence is also a prominent risk factor for various neuropsychiatric illnesses associated with aberrant prefrontal cortex (PFC) functioning, including depression and schizophrenia. We have previously shown that within adolescence, the period of pubertal onset is associated with a number of neuroanatomical changes in the rodent mPFC, including neuron, dendritic, and synaptic pruning. It is hypothesized that the rapidly changing pubertal brain may confer a unique vulnerability to external stressors in adolescents. In support of this, we have previously demonstrated that pubertal stress, but not post-pubertal stress, exerts sex-specific deficits on pre-pulse inhibition and forced swim test performance in adulthood. Additionally, pubertal stress leads to decreased frontal white matter volume in female subjects; this was not the case in adolescent females stressed following the period of pubertal onset. Microglial cells have been implicated in the stress response and play a role in neural development, and recent work suggests that adolescent microglia take on a more activated morphology than adults following an LPS treatment. Moreover, a role for aberrant microglia activation has been implicated in the etiologies of depression and schizophrenia. In this experiment, we exposed male and female rats to a

combination of isolation and restraint stress during the onset of puberty or during the post-pubertal period of adolescence, with an additional unstressed control group. Brain tissue was collected from these subjects in adulthood and immunologically stained for IBA-1, a marker for microglia cells. Microglia were quantified and categorized according to their activation states. We found that in both males and females, neither pubertal nor post-pubertal stress led to long-term changes in microglia numbers or activational states. Future research should focus on more acute changes to microglia number and morphology following stressors in adolescence, which could explain long-term changes in behavior induced by pubertal stress.

Disclosures: C. Drzewiecki: None. **J. Willing:** None. **L. Cortes:** None. **J.M. Juraska:** None.

Poster

372. Insights Into Developmental Vulnerabilities

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 372.11/C59

Topic: A.09. Adolescent Development

Support: NIMH Grant MH100228

Title: Transcriptome and cellular analyses in the human dorsolateral prefrontal cortex: associations with adolescent cannabis use

Authors: *S. MUKHERJEE[1], S. PARK[2], B. GADAD[1], S. PAWAR[1], D. DURAKOGLUGIL[1], I. DOZMOROV[3], K. GLEASON[1], C. TAMMINGA[1], T. HWANG[2], S. GHOSE[1]
[1]Dept. of Psychiatry, [2]Dept. of Clin. Sci., [3]Immunol., UT Southwestern Med. Ctr., Dallas, TX

Abstract: While the acute use of cannabis negatively impacts cognitive function, the long term cognitive effects are controversial. When cannabis use is initiated during adolescence, however, the findings are more consistent with several studies reporting associations between adolescent cannabis use (ACU) and persistent neuropsychological impairment. To determine the molecular and cellular underpinnings of these potential long term effects of ACU, we assembled a cohort of human post mortem brain tissue collected from individuals with a history of adolescent cannabis users (ACU+) and compared them to a cohort of non-cannabis users (ACU-; n=10/group). We conducted whole transcriptome sequencing followed by ingenuity pathway analysis and WGCNA R-package analyses. In addition, we quantified pyramidal neuron somal areas in Layer 3 and Layer 5 in 3 sections of the DLPFC (n=5/group; 80-100 cells//case). About 2000 genes were differentially expressed between ACU+ and ACU- groups. After correcting for multiple comparisons, only 3 genes were significantly different between groups, namely HSPA6, HSPA7 and SERPINA3 (FDR 0.018 to 0.048). Two of these genes belong to the heat shock protein 70 (HSP 70) group and all 3 genes are known to be involved in protein folding and unfolding, processes by which proteins acquire three dimensional structures essential to their biologic

activity. Our network analyses identified differential expressed genes involved in maintaining cellular morphology. Cellular analyses show that layer 3 pyramidal neuronal somal sizes were significantly larger in ACU+ (235.8 $\pm$ 12.4 μm^2) compared to ACU- (186.7 $\pm$ 8.5 μm^2) groups (p=0.01). There were no differences in layer 5. These data suggest that adolescent cannabis use leads to persistent changes in HSP 70 genes which are typically expressed in response to stressors that damage proteins, cause protein unfolding and aggregation. Hsp70 prevents protein aggregation, allows refolding and participates in the disposal of damaged proteins. The increased expression of HSPA6, HSPA7, and SERPINA3 in the ACU+ group could be a compensatory response to potentially deleterious effects of ACU on proteostasis. The differences in genes related to cellular morphology and the increase in layer 3 pyramidal neuronal somal size could reflect a primary process by which ACU increases the neuronal size or could be related to the compensatory responses in an effort to maintain normal cellular physiology in the ACU+ group. These post mortem brains tissue studies are descriptive but do suggest that cannabis use during adolescence leads to persistent molecular and cellular sequelae in the human brain.

Disclosures: S. Mukherjee: None. **S. Park:** None. **B. Gadad:** None. **S. Pawar:** None. **D. Durakoglugil:** None. **I. Dozmorov:** None. **K. Gleason:** None. **C. Tamminga:** None. **T. Hwang:** None. **S. Ghose:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.01/C60

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: NICHD intramural research program

Title: Identification of cell type-specific ErbB4 isoform expression using a novel sensitive *In situ* hybridization approach

Authors: *L. M. ERBEN[1,2], M.-X. HE[3], A. LAEREMANS[3], E. PARK[3], A. BUONANNO[1]
[1]Section on Mol. Neurobiology, NICHD, NIH, Bethesda, MD; [2]Inst. of Mol. Psychiatry, Univ. of Bonn, Bonn, Germany; [3]Advanced Cell Diagnostics, Newark, CA

Abstract: Alternative splicing is ubiquitous in humans, particular prevalent in the central nervous system (CNS) and aberrant splicing is frequently observed in cancers and a hallmark of the psychiatric diseases schizophrenia (SZ). Among the many genes aberrantly spliced in SZ are the neurotrophic factor Neuregulin-1 (NRG-1) and its cognate receptor tyrosine kinase ErbB4. Both *NRG-1* and *ERBB4* polymorphisms are well known for their association with schizophrenia. Four ErbB4 isoforms are generated by alternative splicing of exons in the juxtamembrane (either JMa or JMb variants) and cytoplasmic regions (either CYT-1 or CYT-2

variants) with distinct processing and signaling properties. Several studies reported both the cleavable JMa ErbB4 isoform and the CYT-1 isoform to be increased in postmortem dorsolateral prefrontal cortex of SZ patients. ErbB4 isoform expression is also altered in different brain cancers (e.g. medulloblastoma, astrocytic glioma).
However, the analysis of the expression of ErbB4 isoforms has been limited to quantitative real-time PCR disregarding pivotal information about the regional distribution and cell-type. Here, we analyze ErbB4 isoform expression in the adult mouse brain using a novel sensitive non-radioisotopic *in situ* hybridization (ISH) approach. First, using ErbB4 mutant mice lacking a single exon as controls, we demonstrate that a single pair of paired probes targeting about 50bp at exon junctions provides sufficient sensitivity and specificity to detect exon-exon boundaries. We go on to show that ErbB4 isoforms are differentially expressed in the adult mouse brain. Consistent with our data from quantitative real-time PCR, JMb and CYT-2 are the major ErbB4 isoforms expressed in the mouse brain (e.g. in the hippocampus and cortex). However in the corpus callosum and thalamus, JMa and to a lesser extent CYT-1 are the major ErbB4 isoforms. By combining this new ISH approach with transgenic mice and post-hoc immunohistochemistry, we show that these differences are due to cell type-specific expression of juxtamembrane isoforms. While GABAergic neurons mainly express JMb, oligodendrocytes mostly if not exclusively express the JMa isoform. Our study provides a valuable tool to analyze single-exon splice variants in the brain and other tissue including archival and patient tissue. Further, it highlights novel insights in the function of ErbB4 in oligodendrocytes and the potential implication of this cell type in SZ.
This work was kindly supported by the Eunice Kennedy Shriver NICHD Intramural Research Program, NIH.

Disclosures: L.M. Erben: None. **M. He:** A. Employment/Salary (full or part-time):; employed by ACD. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock owner at ACD. **A. Laeremans:** A. Employment/Salary (full or part-time):; employed by ACD. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock owner at ACD. **E. Park:** A. Employment/Salary (full or part-time):; employed by ACD. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock owner at ACD. **A. Buonanno:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.02/C61

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: The Eunice Kennedy Shriver National Institute of Child Health and Human Development Intramural Research Program

Title: Neuronal processing and axonal targeting of neuregulin 3

Authors: *D. VULLHORST, T. AHMAD, A. BUONANNO
Section on Mol. Neurobio., NICHD, NIH, Bethesda, MD

Abstract: The Neuregulin/ErbB4 signaling pathway is an important modulator of GABAergic inhibitory interneurons, synaptic plasticity and neural network synchronization. Our recent studies indicate that NRGs with a single transmembrane (TM) domain (i.e. NRG1 types I/II and NRG2) accumulate as unprocessed pro-forms at somatic subsurface cistern (SSC)-type ER-PM junctions, where they can signal in paracrine or autocrine fashion following their activity-dependent release. On the other hand, earlier studies showed that the dual-pass transmembrane neuregulin CRD-NRG1 targets to axons and presynaptic terminals where it signals in juxtracrine mode. While NRG3 constitutes one of the most highly expressed NRGs during pre- and postnatal development and has attracted attention for its association with psychosis and its role in interneuron migration, little is known about its subcellular distribution and signaling mode. Using different molecular and cellular biological approaches in cultured hippocampal neurons, here we show that NRG3, like CRD-NRG1, is a dual-pass TM protein. Moreover, NRG3 is also trafficked to axons where it signals in juxtacrine mode via ErbB4 receptors located on dendrites of GABAergic interneurons. In contrast to single-pass TM NRGs that are processed by metalloproteinases on the cell surface, both NRG3 and CRD-NRG1 are processed by BACE in the cell body. In fact, BACE-mediated processing is a prerequisite for axonal accumulation of NRG3, as interference with processing by pharmacological or genetic means causes accumulation of the pro-form in the trans-Golgi network. Lastly, live-imaging experiment indicate that axonal trafficking of processed NRG3 occurs via Rab4 containing vesicles. These results reveal previously unknown molecular mechanisms underlying NRG3 processing and trafficking in neurons, which has important implications for its biological functions and possible involvement in psychiatric disorders. Furthermore, taken together with our work on single-pass NRG isoforms, these findings demonstrate that NRG isoforms can be classified into two major groups based on their distinct transmembrane topologies and their subcellular distribution that likely regulate neuronal functions in fundamentally different ways.
Support: The Eunice Kennedy Shriver National Institute of Child Health and Human Development Intramural Research Program.

Disclosures: D. Vullhorst: None. **T. Ahmad:** None. **A. Buonanno:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.03/C62

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: Intramural Research Program of the Eunice Kennedy Shriver NICHD to M.S., I.K., A.S., L.E., D.V, and A.B.

Intramural Research Program of the NIMH to J.G-O., and S.A.

Intramural Research Program of the NINDS to J-H.S., and V.A.

Title: Direct neuregulin-ErbB4 signaling in dopaminergic axonal projections regulates extracellular dopamine levels and cognitive-related behaviors

Authors: *M. SKIRZEWSKI[1], I. D. KARAVANOVA[2], A. SHAMIR[3], L. M. ERBEN[4], J. GARCIA-OLIVARES[5], J. SHIN[6], D. VULLHORST[7], V. A. ALVAREZ[8], S. G. AMARA[9], A. L. BUONANNO[10]
[1]Section Mol. Neurobio., NIH/NICHD, Bethesda, MD; [2]NIH-NICHD, Bethesda, MD; [3]Mazra Mental Hlth. Ctr., Akko, Israel; [4]Section on Mol. Neurobiology, NICHD, NIH, Bethesda, MD; [5]Natl. Inst. of Mental Hlth., Bethesda, MD; [6]NIAAA, The Natl. Inst. of Hlth., Bethesda, MD; [7]Section on Mol. Neurobio., NICHD, NIH, Bethesda, MD; [8]Lab. of Integrative Neurosci., Natl. Inst. on Alcohol Abuse and Alcoholism, Bethesda, MD; [9]Natl. Inst. of Mental Hlth., Bethesda, MD; [10]Head of Cell Biol. Affinity Group, NICHD, Chief of Sect Mol Neurobiol (SMN), NICHD, NIH, Bethesda, MD

Abstract: Single nucleotide polymorphisms in genes for Neuregulin 1 (NRG1) and its receptor ErbB4 confer susceptibility for schizophrenia, a neurodevelopmental disorder characterized by dopamine (DA) dysfunction and excitatory/inhibitory imbalance. ErbB4 is abundantly expressed in parvalbumin (PV)+ interneurons in the hippocampus and cortex, and in midbrain tyrosine hydroxylase (TH)+ neurons. Although NRG/ErbB4 signaling has been reported to regulate synaptic plasticity, oscillatory activity and behaviors relevant in psychiatric disorders, it presently is unclear relative the contribution of ErbB4 signaling in either PV+ or dopaminergic (DAergic) neurons in regulating these functions. Prior studies reported that local administration of NRG1 by reverse microdialysis into the dorsal hippocampus of freely moving rats results in a rapid increase of extracellular DA levels (Kwon et al, 2008), suggesting that NRG1-ErbB signaling regulates extracellular DA levels either by directly modulating DAergic neurons or by indirectly regulating inhibition of DAergic processes. Here we provide neurochemical, biochemical and behavioral evidence demonstrating that ErbB4 signaling in DAergic axonal projections directly regulate *in vivo* DA homeostasis in brain areas reportedly to be affected in

psychiatric disorders, such as hippocampus, medial prefrontal cortex and striatum. Additionally, we used Lund Human Mesencephalic (LUHMES) cells, an immortalized DAergic cell line that releases DA and express ErbB4, to show that ErbB4 activation also regulates extracellular DA levels by reducing DAT-dependent uptake. Interestingly, mice lacking ErbB4 in TH+ neurons exhibit a dual DA imbalance reminiscent to schizophrenia and cognitive-related behavioral deficits that can be ameliorated if ErbB4 is specifically reintroduced in midbrain TH+ neurons. Based in our findings, it is tempting to suggest that studies exploring the role of the NRG-ErbB4 signaling in excitatory/inhibitory balance should not limit their interpretation to effects originating exclusively from ErbB4-expressing GABAergic interneurons, but also should consider the neuromodulatory effects of DAergic signaling. *This work was kindly supported by the Eunice Kennedy Shriver NICHD Intramural Research Program, NIH.*

Disclosures: M. Skirzewski: None. **I.D. Karavanova:** None. **A. Shamir:** None. **L.M. Erben:** None. **J. Garcia-Olivares:** None. **J. Shin:** None. **D. Vullhorst:** None. **V.A. Alvarez:** None. **S.G. Amara:** None. **A.L. Buonanno:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.04/C63

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: Intramural Research Program of the Eunice Kennedy Shriver NICHD.

Title: Neuregulin-2 regulates dopamine homeostasis, NMDA receptor synaptic currents and behaviors relevant to psychiatric disorders

Authors: *A. L. BUONANNO[1], L. YAN[2], A. SHAMIR[3], E. LEIVA-SALCEDO[4], O.-B. KWON[5], I. D. KARAVANOVA[6], D. PAREDES[7], O. MALKESMAN[8], K. R. BAILEY[9], J. N. CRAWLEY[10], D. VULLHORST[11], M. SKIRZEWSKI[12]
[1]Head of Cell Biol. Affinity Group, NICHD, Chief of Sect Mol Neurobiol (SMN), NICHD, NIH, Bethesda, MD; [2]Section of Mol. Neurobio., NICHD, Bethesda, MD; [3]Mazra Mental Hlth. Ctr., Akko, Israel; [4]Dept. de Biologia, Univ. de Santiago, Santiago, Chile; [5]DGMIF, Daegu, Korea, Republic of; [6]NIH-NICHD, Bethesda, MD; [7]Lieber Inst. /John Hopking Univ., Baltimore, MD; [8]HHMI, Ashburn, VA; [9]Psychology, Susquehanna Univ., Selinsgrove, PA; [10]Dept. of Psychiatry and Behavioral Sci., Univ. of California Davis Sch. of Med., Sacramento, CA; [11]Section on Mol. Neurobio., NICHD, NIH, Bethesda, MD; [12]Section Mol. Neurobio., NIH/NICHD, Bethesda, MD

Abstract: Prior studies have associated polymorphisms in the genes encoding Neuregulin-1 (NRG1) and its receptor ErbB4 with endophenotypes relevant to psychiatric disorders, including schizophrenia. Although the role of NRG1 in neurophysiological and behavioral phenotypes has

been extensively studied, little is known about the function of NRG2 - the closest NRG1 homologue. NRG2 transcript levels increase postnatally after birth in brain areas that are affected in psychiatric disorders, such as the striatum and medial prefrontal cortex (mPFC), while NRG1 expression is widespread in the embryonic and fetal brain but becomes more restricted during adulthood. We therefore generated NRG2-KO mice to investigate to what extent signaling via this ligand accounts for the phenotypes observed in ErbB4 mutant mice that include an imbalance in extracellular dopamine (DA) levels, altered synaptic plasticity and numerous behavioral deficits relevant to psychiatric disorders (Shamir et al. JNS 2012). Using *in vivo* microdialysis, we found that NRG2-KO mice show high extracellular DA levels in dorsal striatum but low levels in the mPFC - a regional DA imbalance that is similar to that reported for schizophrenia subjects. Moreover, like ErbB4-KO mice, NRG2-KOs showed altered performance in a battery of behavioral tasks requiring striatal and mPFC function, including deficits in working memory during T-maze. Interestingly, acute systemic administration of the atypical antipsychotic clozapine in NRG2-KO mice ameliorated working memory deficits, and the improvement in working memory coincided with a rapid increase in extracellular DA levels in the mPFC. In addition to the altered DA function, NRG2-KO mice also showed augmented NMDA receptor synaptic currents at hippocampal glutamatergic synapses sensitive to ifenprodil, suggesting an increased contribution of GluN2B-containing NMDA receptors. Together, our findings point to a novel role for NRG2 in the adult rodent brain regulating excitatory/inhibitory balance potentially modulated by DA and behaviors with relevance to psychiatric disorders. *This work was kindly supported by the Eunice Kennedy Shriver NICHD Intramural Research Program, NIH.*

Disclosures: **A.L. Buonanno:** None. **L. Yan:** None. **A. Shamir:** None. **E. Leiva-Salcedo:** None. **O. Kwon:** None. **I.D. Karavanova:** None. **D. Paredes:** None. **O. Malkesman:** None. **K.R. Bailey:** None. **J.N. Crawley:** None. **D. Vullhorst:** None. **M. Skirzewski:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.05/D1

Topic: B.01. Neurotransmitters and Signaling Molecules

Title: An ErbB antagonist injected into the substantia nigra decreases dopamine release in the striatum and inhibits feeding behavior

Authors: M. M. RAMIREZ[1], *L. R. BETANCOURT[2,1], D. PAREDES[3], L. F. HERNANDEZ[1,4], P. RADA[1]
[1]Human Physiol., [2]Morphological Sci., Univ. of Los Andes, Libertador, Venezuela, Bolivarian

Republic of; [3]Knoebel Inst. for Healthy Aging, Univ. of Denver, Denver, CO; [4]Miller Sch. of Med., Univ. of Miami, Miami, FL

Abstract: The dopaminergic system is linked to locomotor as well as motivated behaviors including feeding and reward. On the other hand, ErbB4 receptors that bind neuregulin-1 (NRG1) are expressed, among other brain sites, in dopamine (DA) and GABA neurons in the midbrain and are involved in myelin formation, axon migration and neuronal survival. Moreover, injection of neuregulin-1 directly in the substantia nigra (SN) increases DA release in the corpus striatum (STR) confirming a link between neuregulin-ErbB4 and the neurotransmission process. In the present report, we have monitored DA release in the STR, following injection of the ErbB receptor tyrosine kinase inhibitor PD158780 (PD15) in the SN. In a separate set of rats, PD15 was injected in the SN and free feeding was monitored during the active and inactive circadian cycle of the rat. Finally, the effect of PD15 in the SN on the hedonic value of a palatable food was tested using the sucrose preference test. The injection of PD15 (80 nMoles) into the SN decreased DA levels in the STR to 56.7 ± 10% ($F(7,91)=2.946, p<0.01$). This was correlated with a significant decrease in food chow consumption. Rats injected PD15 at the beginning of the active cycle ate significantly less (0 ± 0 g during the first hour and 5 ±1.5 g after 4 hr) than control animals (5.9 ± 2 g at the hour and 10.7 ± 2.1 after 4 hr) ($T(10)=3.219$, $p<0.01$ 1st hr and $T(10)=2.371$, $p<0.05$ 4hr total). PD15 injected at the beginning of the inactive cycle also decreased food ingestion (1.1 ± 0.6 g and 3 ± 0.6 g, 1-hr and 4-hr respectively in PD15 compared to 2.4 ± 0.6 g and 6.3 ± 1.2 g 1-hr and 4-hr post injection in controls ($T(10)=1.558$; N.S. for 1st hr, $T(10)= 2.545$, $p<0.05$ for 4hr total). Drinking was not significantly affected by injection of PD15 in the SN neither did it affect the sucrose preference index. These results show that ErbB4 receptors in the SN are not only important in DA-neuron development and survival, but are also involved in directly modulating DA neurotransmission in the STR as supported by a decrease in DA release when the ErbB4 antagonist was injected. This modification is correlated with a change in feeding suggesting that normal function of the nigrostriatal in ingestive behavior is dependent, among other systems, on neuregulin-ErbB4. This could be useful in finding new therapeutic approaches in the treatment of obesity.

Disclosures: M.M. Ramirez: None. **L.R. Betancourt:** None. **D. Paredes:** None. **L.F. Hernandez:** None. **P. Rada:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.06/D2

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: UIC start-up funds

Title: TrkB regulates activity-dependent translocation of ZDHHC8 to dendrites through its phosphorylation by PKC

Authors: *A. YOSHII[1], K. P. KOSTER[2], Y. MURATA[3]
[1]Dept of Anat. & Cell Biol., UIC, Chicago, IL; [2]Dept. of Anat. and Cell Biol., Univ. of Illinois at Chicago, Chicago, IL; [3]Pharmacol. and Physiol., George Washington Univ., Washington, DC

Abstract: Post-synaptic density protein-95 (PSD-95) is a major scaffolding protein for ionotropic glutamate receptors and signaling molecules. It mediates many forms of synaptic plasticity and is critical for brain maturation, learning, and memory. In the rodent visual pathway, PSD-95 becomes redistributed to synapses within a few hours of eye-opening at postnatal day 13 when the patterned vision starts. The Increase in synaptic activities activates brain-derived neurotrophic factor (BDNF) and its receptor TrkB signaling pathway, and regulates the vesicular transport of PSD-95 to synapse. Attachment of PSD-95 to the cargo membrane requires palmitoylation of the protein. This lipid modification also facilitates proper subcellular localization of a modified protein to a membranous microdomain, lipid raft. PSD-95 palmitoylation is critical for synaptic connections and refinements. However, the regulatory mechanisms are largely unknown. Previously, we showed that palmitoylation of PSD-95 is regulated by BDNF through phospholipase C (PLC) and a PKC variant. We also identified the palmitoylation enzyme ZDHHC8 as the target of PKC phosphorylation but not other members of ZDHHC family. This pathway indeed regulated trafficking of PSD-95 to synapses in developing brains *in vivo*. Here, we used a phospho-proteomic approach, and identify phosphorylation sites of ZDHHC8. We generated mutated ZDHHC8 constructs by targeting the phosphorylation sites. We performed immunoprecipitations to directly assess the mutant proteins using an antibody specific to a PKC phosphorylation motif, and confirmed that one of the mutations are regulated by a PKC. In cultured neurons, the mutant PKC mutant ZDHHC8 localized in the Golgi apparatus while the intact enzyme was distributed throughout dendrites. In layer 2/3 pyramidal neurons of the visual cortex, eye-opening trigger ZDHHC8 trafficking from soma to dendrites *in vivo*. However, the phosphorylation-inactivated mutant ZDHHC8 remained predominantly in the soma even after eye-opening. Inhibition of TrkB suppresses PSD-95 trafficking to visual cortical synapses upon eye opening. An overexpression of the intact but not the mutant ZDHHC8 rescued the synaptic redistribution of PSD-95. Therefore, TrkB regulates the PKC phosphorylation of ZDHHC8 and facilitates palmitoylation of PSD-95. This study is the first complete example of how a receptor signaling pathway can regulate phosphorylation of a palmitoylation enzyme, thereby triggering protein palmitoylation and synaptogenesis. Our findings have broad implications for various brain disorders in both children and adults.

Disclosures: A. Yoshii: None. **K.P. Koster:** None. **Y. Murata:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.07/D3

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: ERC grant No 322742 – iPLASTICITY

Academy of Finland

Sigrid Juselius foundation

Title: TrkB deletion from serotonergic neurons leads to impairment in memory and antidepressant efficacy

Authors: *M. PRIYADARSHINI SAHU[1], A. STEINZEIG[2], O. BOROWIECKI[2], V. VOIKAR[2], E. CASTREN[3]
[1]Univ. of Helsinki, Helsinki, Finland; [2]Neurosci. Center, Univ. of Helsinki, Helsinki, Finland; [3]Neurosci. Ctr., Helsinki, Finland

Abstract: Antidepressants mainly the selective serotonin reuptake inhibitors (SSRI) drugs activate neurotrophin signaling and neuronal plasticity. Brain derived neurotrophic factor (BDNF) together with its high affinity cognate receptor, tyrosine kinase B (TrkB) plays a significant role in neuronal survival and synaptic plasticity and have been concomitantly related with serotonin (5-HT) in a myriad of neurochemical and behavioral responses with SSRI treatment. The mechanism of this interplay between BDNF/TrkB with 5-HT is limited. To investigate the interaction between these signalling molecules, we have used a conditional knockout for TrkB receptors from tryptophan hydroxylase2 (Tph2) producing serotonergic neurons in the mice. The inducible cre-lox recombination system was used by injecting tamoxifen to six weeks old mice. These knockout mice have been characterized in a series of behavior tests related to depression and anxiety and observed increased hyperactivity. We further investigated antidepressant efficacy of fluoxetine in the knockouts. The knockouts responded to acute effects of fluoxetine but the chronic effect was attenuated. There was no signs of aggression in the knockouts using the resident intruder paradigm. We monitored the metabolic changes in these mice, using CLAMS (Comprehensive lab animal monitoring system). The knockouts were leaner in comparison to the control littermates while no change in feeding behavior was observed. Furthermore, we assessed the learning and memory in these mice by contextual fear conditioning. The results suggest impairment in memory consolidation in the knockouts. At the cellular and molecular levels, serotonin levels in different brain regions have been measured and was found to be augmented. Thus impairment of TrkB signalling in the Tph2 neurons in early adulthood leads to disturbances in the 5-HT neurotransmission and BDNF

signalling. These functional deficits contribute to blunted memory consolidation and response to SSRI. Further studies on the downstream signalling cascades and serotonin receptors could possibly shed more information on the critical balance between the two systems that contribute to neuronal development, synapse formation and plasticity.

Disclosures: **M. Priyadarshini Sahu:** None. **A. Steinzeig:** None. **O. Borowiecki:** None. **V. Voikar:** None. **E. Castren:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.08/D4

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: NIH Grant NS051710-11

NIH Grant NS083057

American Epilepsy Society

CURE Epilepsy

Title: Whole transcriptome analysis of the BDNF-induced JAK/STAT response in neurons and in a TLE model of epilepsy

Authors: ***K. M. HIXSON**[1], M. COGSWELL[2], A. BROOKS-KAYAL[3], S. J. RUSSEK[4]
[1]BUSM Pharmacol., Dorchester, MA; [2]Pharmacol. and Exptl. Therapeut., Boston Univ., Boston, MA; [3]Neurol., Children's Hosp, Aurora, CO; [4]Pharmacol. and Grad Prog for Neurosci., Boston Univ. Sch. Med., Boston, MA

Abstract: Deleterious alterations in molecular signaling cascades and cellular remodeling initiate years prior to the onset of temporal lobe epilepsy (TLE). Despite many upstream cellular and molecular changes, most drugs only target neurotransmission to increase inhibition. Over 30% of patients are refractory to all of these medications. Understanding the molecular basis of acquired epilepsy and developing efficacious drugs to target intracellular signaling pathways is vital in the advancement of patient therapy. Upregulation of brain-derived neurotrophic factor (BDNF) is long established as a contributing factor to epilepsy development in humans and animal models. More recently, our lab discovered that BDNF induces activation of the Janus Kinase/Signal Transducer and Activator of Transcription (JAK/STAT) pathway in neurons and its inhibition attenuates spontaneous seizures in a TLE animal model. In this study, we elucidate key gene expression changes involved in the BDNF-induced JAK/STAT pathway through deep RNA-sequencing of primary cortical neurons treated with different concentrations of the

JAK/STAT inhibitors, WP1066 and Ruxolitinib. This work has led to the development of a molecular fingerprint to investigate alterations in the transcriptome that occur in transgenic mice engineered to conditionally knockdown expression of STAT3 as well as the BDNF receptor, p75NTR, two proteins that form intracellular complexes in neurons and are associated with TrkB mediated activation of JAK/STAT pathway. Of great interest to us is the observation that the genes most associated with BDNF-induced responses that are blocked by JAK inhibitors are mapped to neurological diseases, with the top four categories being relevant to epileptic seizures and epilepsy. In addition, both classes of inhibitors target the expression of type A GABA receptor subunits as well as Scn1A, a gene implicated in early onset epileptic encephalopathies such as Dravet Syndrome. The emerging landscape of the transcriptome for BDNF-induced JAK/STAT signaling suggests that it may be useful for early-detection biomarkers that inform seizure susceptibility as well as contain new targets for the future treatment of intractable epilepsies.

Disclosures: K.M. Hixson: None. **M. Cogswell:** None. **A. Brooks-Kayal:** None. **S.J. Russek:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.09/D5

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: NIH Grant MH100652

NIH Grant MH053851

Title: Effects of the JAK2/STAT3 activator CNTF on Arc expression in the rat orbitofrontal cortex

Authors: *M. GIROTTI[1], D. A. MORILAK[2]
[1]Pharmacol., [2]Pharmacol. and Ctr. for Biomed. Neurosci., Univ. of Texas Hlth. Sci. Ctr. at San Antonio, San Antonio, TX

Abstract: Cognitive flexibility is the ability to modify established behaviors or previous learning in response to a change in the environment, for example, by a change in stimulus-reward contingency. Reversal learning is a specific form of cognitive flexibility predominantly mediated by the orbitofrontal cortex (OFC), and is impaired in a range of psychiatric conditions, including depression and obsessive compulsive disorder. In previous work, we reported that basal activity of the JAK2/STAT3 signaling pathway in the OFC is required for optimal performance on a reversal learning task under healthy control conditions. Ciliary neurotrophic factor (CNTF) is an

endogenous activator of JAK2/STAT3 signaling in the brain, and is known to exert pro-survival effects on both neurons and oligodendrocytes. In preliminary studies using primary neuronal cultures, we found that CNTF induced the synaptic plasticity protein Arc. This suggested that CNTF could potentially influence neuronal plasticity underlying reversal learning in the OFC. Indeed, we found that blocking endogenous CNTF with an intra-OFC microinjection of a neutralizing CNTF antibody was sufficient to produce reversal-learning deficits. In the current work, we present evidence that the local microinjection of CNTF induces the phosphorylation of both JAK2 and STAT3 in the OFC in a dose dependent manner, but with slightly different profiles. While both JAK2 and STAT3 were induced by 100 nM CNTF, only JAK2, but not STAT3, was activated by 50 nM CNTF. Arc expression also was induced by CNTF in the OFC. Interestingly, the CNTF dose profile of Arc expression followed that of JAK2 activation, peaking at 50 nM and actually declining at 100 nM CNTF, suggesting the possibility that JAK2 is specifically involved in modulating Arc levels. Supporting this hypothesis, in cultured neurons, we have found that silencing JAK2 but not STAT3 reduces Arc expression.
Arc is specifically expressed in glutamatergic pyramidal neurons. Using immunohistochemistry we have found that CNTF induces JAK2 in CamK-II positive excitatory neurons. Thus, we hypothesize that CNTF, at low concentrations, increases Arc expression in pyramidal neurons by a JAK2-dependent mechanism, and that this phenomenon is required for optimal reversal learning. In future work we will test this hypothesis using a viral vector containing a CamK-II driven silencing hairpin RNA against JAK2. We expect that knocking-down JAK2 in glutamatergic neurons will reduce Arc expression and produce deficits in reversal learning.
Support: NIH grant MH100652 and MH053851.

Disclosures: **M. Girotti:** None. **D.A. Morilak:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.10/D6

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: NS21072

Title: Genetic variants in scaffold proteins ARMS/KIDINS220 and RHO-GEF TRIO lead to altered plasma membrane expression and decreased dendritic transport in primary cortical neurons

Authors: *T. M. KRANZ[1], A. FOMITCHOVA[1], D. KAMINSKI[1], M. CAMMER[2], A. HEGUY[3], R. C. FROEMKE[1], D. MALASPINA[4], M. V. CHAO[1]
[1]Skirball Inst. of Biomolecular Med., New York Univ. Med. Ctr., New York, NY; [2]Microscopy

Core, Office of Collaborative Sci., [3]Genome Technol. Ctr., New York Univ. Med. Ctr., New York City, NY; [4]Dept. of Psychiatry, New York Univ. Med. Ctr., New York, NY

Abstract: Multiple genes and their pleiotropic interactions are involved in the etiology of complex neuropsychiatric disorders such as schizophrenia. Although many genes have been identified, the signaling pathways have not been fully understood. In this study, we investigated two of these genes, the Ankyrin Repeat-rich Membrane Spanning Protein/Kinase D-Interacting Substrate of 220 kDa (*ARMS/Kidins220)* and the TRIO Rho guanine nucleotide exchange factor (*TRIO*). Both genes harbored missense coding mutations and rare polymorphisms, which were discovered by targeted exome capture in schizophrenia-related psychosis cases. The ARMS/Kidins220 gene displayed a novel mutation in its proline-rich domain (H1085R) that has not been observed before. ARMS/Kidins220 is a scaffold protein involved in neurotrophin signaling and a hub for multiple signaling pathways. TRIO harbored one rare missense coding variant (K3037M) and one novel mutation in its C-terminal kinase domain (V2941I). TRIO is a RHO-GEF involved in cytoskeletal organization and cell migration and an interaction partner of ARMS/Kidins220. Here, we created mutants of ARMS/Kidins220 and TRIO using site-directed mutagenesis and overexpressed the wildtype constructs in comparison to the mutants in HEK293 cells and primary cortical neurons from rats. Using biotinylation, overexpressed ARMS/Kidins220 H1085R yielded less cell surface expression in comparison to the wildtype control in HEK293 cells. Ectopic expression of the ARMS/Kidins220 H1085R in primary rat cortical neurons resulted in impaired transport of the mutant ARMS/Kidins220 protein from soma into neuronal projections. In contrast, the TRIO variants (V2941I and K3037M) did not alter mutant protein localization. We conclude that missense coding variants in ARMS/Kidins220 and TRIO have demonstrable effects on neuronal phenotypes and may provide a potential influence in neuropsychiatric disorders through their interaction.

Disclosures: T.M. Kranz: None. **A. Fomitchova:** None. **D. Kaminski:** None. **M. Cammer:** None. **A. Heguy:** None. **R.C. Froemke:** None. **D. Malaspina:** None. **M.V. Chao:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.11/D7

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: DBT Grant No. BT/PR/5033/MED/30/797/2012

Title: Role of Nerve growth factor and Brain derived neurotrophic factor in regulation of central nervous system myelination

Authors: *J. M. LANGHNOJA, L. BUCH, P. PILLAI
Zoology, The Maharaja Sayajirao Univ. of Baroda, Vadodara, India

Abstract: Neurotrophins are a family of proteins essential for the development and regulation of Central Nervous System (CNS) and are widely implicated as potential modulator for important CNS functions such as neural stem cells differentiation, cell migration, myelination and synaptic plasticity. Neurotrophins comprises of four structurally related factors: brain derived neurotrophic factor (BDNF), nerve growth factor (NGF), neurotrophin-3 (NT-3) and neurotrophin-4/5 (NT-4/5). In CNS myelination, oligodendrocytes form a stable and rigid layer of tightly packed lipid and protein layer known as myelin sheath which is wrapped around the axon of neurons. The oligodendrocytes undergo a specific lineage involving migration, proliferation and ultimately differentiating into the myelinating oligodendrocytes. In adult, oligodendrocytes also differentiate from Neural Stem Cells (NSCs). This lineage progression and differentiation is supported by a number of extracellular cues like growth factors- PDGF and neurotrophins from the surrounding environment. NGF, a major neurotrophin is known to be involved in promoting growth, survival, and stabilization of primary afferents neurons and neurons axonal dendrites along with the differentiation of NSCs into neurons. BDNF, another vital neurotrophin, plays an important role in the initiation of the CNS myelination and stimulation of migration of cerebellar granule cells and Schwann cells. Neurotrophins also support for survival of myelinating cells and remyelination during inflammation and in chronic injury conditions where the myelin proteins' level goes down. However, the signaling mechanisms underlying the molecular action of neurotrophins in the differentiation of NSCs, oligodendrocyte migration and lineage progression along with their role in remyelination is still not well understood. Present study indicates important role of these neurotrophins in the differentiation of NSCs into brain cells majorly into Myelin Basic Protein (MBP) positive oligodendrocytes via the ERK pathway. Moreover, NGF and BDNF also enhance Oligodendrocytes precursor cells' (OPCs) migration; this action being mediated by specific neurotrophins' receptors. Up-regulation of MBP expression in TNF-α induced demyelination shows remyelination potential of neurotrophins in CNS myelination. Overall, our study reveals that NGF and BDNF regulate CNS myelination by NSC differentiation, OPCs migration and may be remyelination in TNF-α induced demyelination.

Disclosures: J.M. Langhnoja: None. **L. Buch:** None. **P. Pillai:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.12/D8

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: Provost's Office for Research (PRP) of University of São Paulo for Center for Advanced Studies in Rehabilitation

São Paulo Research Foundation (FAPESP)

National Council for Research (CNPq) for Daiane Fontes (#131294/2014-8)

Title: Frequency of the BDNF Val66Met polymorphism in Brazilian healthy subjects

Authors: *M. SCHULTZ, D. FONTES, L. S. SANTOS, M. PEDRAZZOLI
Univ. of Sao Paulo, Sao Paulo, Brazil

Abstract: Purpose. The Val66Met single-nucleotide polymorphism (rs6265) of the BDNF gene has been implicated to central nervous system (CNS) dysfunctions by interfering on neurotrophin production and transportation. The present study reports the frequency of BDNF Val66Met in 1284 Brazilian healthy volunteers. Stratification by race was performed to identify patterns of distribution. **Methods.** The total of subjects was 1284 healthy college students, 752 men (48.3%) and 806 women (51.7%), with averaged age of 21.53±4.71, from three regions in Brazil: north (Alagoas), south (Rio Grande do Sul) and southeast (São Paulo). The Ethics Committee of the School of Arts, Sciences and Humanities of University of São Paulo approved the study. The subjects auto-declared their race and were divided in five groups: yellow, white, black, brown and indigenous, according to the Brazilian Institute of Geography and Statistics. The DNA was obtained from buccal epithelial cells. For Val66Met genotyping, the Restriction Fragment Length Polymorphism (RFLP) was applied using the High Resolution Melt (HRM) as tool. PCR amplifications were carried using the primer BDNF-RS6265 (*Invitrogen*, USA). The genotypes Val/Val, Val/Met and Met/Met were determined by the melting curve in the PCR, according to the previous parameters. The Hardy-Weinberg equilibrium was used for genotypic and allelic distribution. **Results.** The genotypic distribution was 66.08% for Val/Val, 30.38% for Val/Met and 3.54% for Met/Met. Allele frequencies were 81% for Val and 19% for Met. Genotype and allelic distributions were similar in men and women. The analysis of genotypic distribution by the race reveals the total of 826 white subjects distributed as follow: 530 (64,2%) Val/Val, 260 (31,5%) Val/Met and 36 (4,4%) Met/Met. The brown race has had the total of 342 subjects: 246 (71,9%) Val/Val, 90 (26,3%) Val/Met and 6 (1,8%) Met/Met. 55 subjects were yellow: 28 (50,9 %) were Val/Val, 23 (41,8%) Val/Met e 4 (7,3%) Met/Met. Black individuals were 56 and the genotypes were: 45 (80,4%) Val/Val and 11 (19,6%) Val/Met. Five subjects were indigenous: 4 (80%) were Val/Val and only 1 (20%) had have Val/Met. None black and indigenous subjects had have Met/Met genotype. **Conclusion.** The present study aimed to characterize the Val66Met in healthy Brazilians and to identify patterns of distribution by the race. Brazilian population is a resultant of cross-cultural relations, which involves migration flux along the history. Even considering an interracial characteristic, our findings confirm a higher occurrence of the Val66Met in yellow race and absent of the polymorphism in black and indigenous people, corroborating to the other studies.

Disclosures: M. Schultz: None. **D. Fontes:** None. **L.S. Santos:** None. **M. Pedrazzoli:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.13/D9

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: the Ministry of Education, Science and Culture of Japan 15K19743

the Ministry of Education, Science and Culture of Japan 15K15432

the Uehara Memorial Foundation

the Osaka Medical Research Foundation for Intractable Disease

the Naito Foundation

the Takeda Science Foundation

Title: Microglia-derived neuregulin expression in psychiatric disorders

Authors: *D. IKAWA, M. MAKINODAN, T. YAMAUCHI, Y. YAMASHITA, T. KOMORI, T. KISHIMOTO
Nara Med. Univ., Kashihara / Nara, Japan

Abstract: Considerable evidence shows that neuregulins (NRGs) are involved in brain function and psychiatric disorders. While NRGs have been regarded as neuron- or astrocyte-derived molecules, our research has revealed that microglia also express NRGs, levels of which are markedly increased in activated microglia. Previous studies have indicated that microglia are activated in the brains of individuals with autism spectrum disorder (ASD). Hence, we investigated microglial NRG mRNA expression in multiple lines of mice considered models of ASD. Curiously, while maternal immune activation (MIA) mice exhibited identical microglial NRG expression to controls, NRG expression significantly increased in the microglia of BTBR and socially-isolated mice compared to those in controls. Furthermore, we observed a positive correlation between NRG expression in microglia and peripheral blood mononuclear cells (PBMCs) in mice. This finding suggests that NRG expression in human PBMCs may mirror microglia-derived NRG expression in the human brain. To translate these findings for application in clinical psychiatry, we measured levels of NRG1 splice-variant expression in clinically available PBMCs of patients with ASD. Levels of NRG1 type III expression in PBMCs were positively correlated with impairments in social interaction in children with ASD (as assessed using the Autistic Diagnostic Interview-Revised test: ADI-R). These findings indicate the possibility that immune cell-derived NRGs may be implicated in the pathobiology of psychiatric disorders such as ASD.

Disclosures: **D. Ikawa:** None. **M. Makinodan:** None. **T. Yamauchi:** None. **Y. Yamashita:** None. **T. Komori:** None. **T. Kishimoto:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.14/D10

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: JSPS KAKENHI Grant 15K12570

Title: Exercise combined with low-level $GABA_A$ receptor inhibition modulates the expression of BDNF in the hippocampus accompanied by changes in epigenetic regulation

Authors: ***H. MAEJIMA**[1], S. NINUMA[2], A. OKUDA[2], T. INOUE[3], M. HAYASHI[3]
[1]Dept. of Rehabil. Science, Fac. of Hlth. Sci., [2]Dept. of Hlth. Sciences, Sch. of Med., [3]Grad. Sch. of Hlth. Sci., Hokkaido Univ., Sapporo, Japan

Abstract: Neurotrophins play a crucial role in neuroplasticity, neurogenesis, and neuroprotection in the central nervous system. Aerobic exercise is known to increase expression of neurotrophins, particularly brain-derived neurotrophic factor (BDNF), in the hippocampus and to improve cognitive function. Several animal studies have evaluated the tonic inhibition of GABAergic synapses to enhance hippocampal plasticity as well as learning and memory. Epigenetic mechanism plays a crucial role to regulate gene transcription. Specifically, the activity levels of histone acetyltransferases (HATs) and histone deacetylases (HDACs) regulate histone acetylation and modulate gene transcription. The objective of the present study was to examine the interactive effect of low-level $GABA_A$ receptor inhibition and exercise on behavior tests (cognitive function and locomotion activity), expression of BDNF and epigenetic regulations, including the activity of HATs and HADCs in the hippocampus. ICR mice were randomly distributed among 4 groups based on two factors of $GABA_A$ receptor inhibition and exercise, i.e. control group, an exercise group, a bicuculline group, and an exercise plus bicuculline group. We administered $GABA_A$ receptor antagonist, bicuculline intraperitoneally to the mice (bicuculline and exercise plus bicuculline group) at a non-epileptic dose of 0.25 mg/kg, whereas the mice (exercise and exercise plus bicuculline group) were exercised on a treadmill for 1 h a day, 5 days a week. After four week intervention, the expression of mRNA and protein abundance of BDNF, activity of HAT and HDAC in the hippocampus were assayed using real time PCR and ELISA following behavioral tests. All study procedures were approved by the ethics committee for animal research of Hokkaido University in Japan. Although we could not confirm a significant improvement of cognitive function assessed by a novel-object recognition test, exercise in the presence of bicuculline administration increased locomotion activity, indicating that exercise combined with low-level $GABA_A$ receptor inhibition enhanced physical

activity level of the mice. Aerobic exercise increased mRNA expression and protein abundance of BDNF in the hippocampus, accompanied by enhanced HAT activity in epigenetic regulation. Alternatively, bicuculline administration increased HDAC activity in the hippocampus in epigenetic regulation. Altogether, it was expected that exercise combined with low-level $GABA_A$ receptor inhibition beneficially contribute to neuroprotection of the hippocampus accompanied by the up-regulation of BDNF expression and changes in epigenetic regulation.

Disclosures: H. Maejima: None. **S. Ninuma:** None. **A. Okuda:** None. **T. Inoue:** None. **M. Hayashi:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.15/D11

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: NIH Grant R01AG041944

Title: Aging and an immune challenge lead to reduced levels of BDNF and cholinergic receptor mRNAs in hippocampal synapses in a rodent model of delirium

Authors: *N. TANAKA, S. L. PATTERSON
Biol., Temple Univ., Philadelphia, PA

Abstract: Delirium is an acute failure in brain function, characterized by deficits in cognition and attention. Aging increases the risk of delirium after surgery, injury, or infection (~ 50% of 65-year or older patients in hospitals). Delirium is usually temporary, but its occurrence is associated with an increased risk of eventually developing dementia. The molecular pathophysiology of the disorder remains poorly understood, but animal models may provide some clues. 24-month-old Fischer Brown Norway (F344xBN) rats are generally healthy agers with no major physical or cognitive deficits. However, after a peripheral immune challenge (an i.p. injection of *E. coli*), they exhibit an exaggerated and prolonged inflammatory response in the brain. 4 days after infection, although they have physically recovered (no more fever nor loss of appetite), these aging animals show deficits in a long-term memory task (contextual fear conditioning) as well as elevated levels of hippocampal IL-1β protein (Barrientos et al., 2009). In contrast, their younger counterparts (3-month-old) display no deficits in the memory task and normal levels of IL-1β at this time point. Intriguingly, our time-course studies (4 - 21 days after infection) show that the impairments in memory and IL-1β are paralleled by reductions in hippocampal area CA1 theta burst-evoked late-phase long-term potentiation (L-LTP, a memory-related form of long-lasting synaptic plasticity) and in levels of mature brain-derived neurotrophic factor (mBDNF, required for long-term memory and L-LTP). This suggests that

BDNF may play a role in linking activity in the central immune system to alterations in synaptic plasticity. In order to further investigate how age and an infection can compromise the capacity for BDNF-dependent memory-related synaptic plasticity, we are examining plasticity-related mRNAs in hippocampal subcellular fractions. Levels of BDNF mRNA transcripts containing a long 3' untranslated region (thought to be trafficked to dendrites for local protein synthesis) and the protein coding domain (common to all the mRNA isoforms of BDNF) are both reduced in hippocampal synaptoneurosomes of aged animals 4 days after infection. Reduced availability of BDNF at synapses might contribute to the deficits in memory-related synaptic plasticity. Since BDNF is thought to promote cholinergic neuronal function and survival and mounting evidence suggests that cholinergic deficiency may contribute to delirium and dementia, we are also investigating cholinergic receptor mRNAs. We have examined one isoform of the receptor, and the transcript is reduced in hippocampal synaptoneurosomes of these aged *E. coli* animals.

Disclosures: N. Tanaka: None. **S.L. Patterson:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.16/D12

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: NRF-2016R1D1A1B03934438

Ministry of Science, ICT & Future Planning 2231-415

Title: Molecular basis of Obsessive-Compulsive Disorder: Involvement of TrkB/Slitrk5 in corticostriatal dysfunction

Authors: *M. SONG[1], A. LI[2], I. DINCHEVA[4], S. SHMELKOV[5], I. NINAN[6], F. LEE[3]
[1]Korea Brain Res. Inst., Daegu, Korea, Republic of; [2]Sackler Inst. for Developmental Psychobiology, [3]Weill Cornell Med. Col., New York, NY; [4]Columbia Univ., new york, NY; [5]Neurosci. and Physiol., NYU Sch. of Med., New York, NY; [6]Psychiatry, New York Univ. Sch. of Med., New York, NY

Abstract: Obsessive Compulsive Disorder (OCD) displays a substantial heritable component but few specific molecular genetic risk factors have been identified. Knockout mice lacking Slitrk5 expression display an OCD-like phenotype involving pathologic grooming that is responsive to serotonin reuptake inhibitors and corticostriatal dysfunction. To examine whether Slitrk5 function contribute to the genetic risk for OCD, we re-sequenced the complete protein coding sequence of SLITRK5 in a population sample of human OCD subjects. Direct functional testing of identified OCD-specific rare non-synonymous mutations in Slitrk5 found that all mutations

showed impaired synaptogenic activity as well as diminished binding to PTPδ, a trans-synaptic binding partner of Slitrks. These results demonstrate that rare mutations in SLITRK5 contribute to the genetic risk for OCD in human populations. We previously reported that Slitrk5 modulates BDNF-dependent biological responses through direct interaction with TrkB receptors. Under basal conditions, Slitrk5 interacts primarily with PTPδ; however, upon BDNF stimulation, Slitrk5 shifts to cis-interactions with TrkB. Interestingly, genetic facilitation of TrkB signaling rescued repetitive phenotype of Slitrk5 KO mice while deletion of BDNF expression in OCD circuit induced increase in grooming behavior. The networks of genes implicated in OCD remain obscure, however, these findings suggest that BDNF-dependent regulation of TrkB, Slitrk5, and PTPδ interactions at the synapse may mediate proper functioning of key corticostriatal circuits implicated in OCD.

Disclosures: **M. Song:** None. **A. Li:** None. **I. Dincheva:** None. **S. Shmelkov:** None. **I. Ninan:** None. **F. Lee:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.17/D13

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: FAPESP 2011/22844-0

Title: Intrahippocampal injection of Ouabain triggers dendritic branching in neurons and memory improvement in adult rats

Authors: *C. SCAVONE, A. M. M. ORELLANA, J. A. LEITE, P. F. KINOSHITA, A. R. VASCONCELOS, M. M. CARARO, D. Z. ANDREOTTI, L. S. LIMA, G. F. XAVIER, E. M. KAWAMOTO
Univ. of Sao Paulo, Sao Paulo, Brazil

Abstract: Ouabain (OUA) is a well-known endogenous cardiotonic steroid that binds to Na,K-ATPase (NKA) and can trigger the activation of signaling pathways, such as NFκB, through intracellular Ca^{2+} levels oscillation. Particularly in neurons, the NFκB signaling has been described to have an important role in molecular switch from short to long-term memory also regulating PKA/CREB signaling cascade. Furthermore, CREB activation can lead to an increase in BDNF that can modulate AKT and Wnt/β-Catenin signaling pathways. The aim of this study was to verify whether intrahippocampal injection of OUA (10 nM) was able to modulate the main signaling pathways involved in morphological plasticity and memory formation in hippocampus of male adult rats. For the first time in the literature, our study suggests that intrahippocampal injection of OUA 10 nM can activate WNT/β-Catenin signaling pathway as

well as CREB/BDNF, AKT and NFκB leading to important changes in cellular microenvironment that results in increased dendritic branching in CA1 and DG neurons, with spatial reference memory improvement. Therefore, molecular mechanisms trigged by low doses of OUA seems to be a promising therapeutic strategy to improve morphological plasticity and cognition in adult hippocampus.

Disclosures: C. Scavone: None. **A.M.M. Orellana:** None. **J.A. Leite:** None. **P.F. Kinoshita:** None. **A.R. Vasconcelos:** None. **M.M. Cararo:** None. **D.Z. Andreotti:** None. **L.S. Lima:** None. **G.F. Xavier:** None. **E.M. Kawamoto:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.18/D14

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: MH070596

NS088943

MH081935

DA017392

Title: Developmental and synaptic roles for FRS adapter proteins: Implications for neurotrophin and FGF signaling in the hippocampus

Authors: *S. NANDI[1], K. ALVINA[2], P. J. LITUMA[2], P. E. CASTILLO[2], J. M. HEBERT[1]
[1]Departments of Neurosci. and Genet., [2]Dept. of Neurosci., Albert Einstein Col. of Med., Bronx, NY

Abstract: Neurotrophins (NTRs) and FGFs are implicated in a wide spectrum of brain functions including neurogenesis, dendritogenesis, spinogenesis, synaptic transmission, and plasticity. The mechanism of signal transduction by NTRs and FGFs from their respective cell-surface receptors (NTRKs and FGFRs) in mediating these processes is poorly understood. Using the mouse hippocampus as model we assessed the role of the FGF Receptor Substrate (FRS) family (FRS2 and FRS3), one of the first identified intracellular family of adapter proteins, which transduces signal from both NTRKs and FGFRs. Transcripts of one of several *Ntrk* and *Fgfr* genes (*Ntrk2* and *Fgfr1*, respectively), as well as *Frs2*, were strongly expressed throughout the hippocampus. Within the dentate gyrus (DG), *Ntrk2* and *Fgfr1* expressions were particularly robust in the sub-granular zone, while the granule cell layers showed stronger expressions for *Ntrk2* and *Frs2*. Using brain-specific and germline mutants, we found that FRS2 and FRS3 regulate DG

morphogenesis and postnatal neurogenesis, roles attributed to both NTRK2 and FGFR1. Two-photon imaging further showed that FRS adapters are required for dendritic branch and spine formation in DG granule cells, roles that were so far described for NTRK2. Consistent with a synaptic function for FRS adapters, *Frs* mutant mice displayed reduced excitatory synaptic transmission at glutamatergic perforant path inputs onto DG granule cells without significantly affecting presynaptic function as found by electrophysiological recordings in acute hippocampal slices. These observations reveal important roles for FRS adapters during DG development and postnatal neurogenesis. They further suggest maturational/synaptic function for FRS in DG granule cells. We reason that both NTRs and FGFs can utilize FRS in a contextual, receptor-specific manner in the hippocampus. Whether FRS signaling is directly required for these synaptic functions as well as for activity-dependent, long-term plasticity remains to be determined.

Disclosures: S. Nandi: None. **K. Alvina:** None. **P.J. Lituma:** None. **P.E. Castillo:** None. **J.M. Hebert:** None.

Poster

373. Regulation and Function of Neurotrophic Factors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 373.19/D15

Topic: B.01. Neurotransmitters and Signaling Molecules

Support: DC007176

OGMB160167

Title: Blocking BDNF-TrkB signaling reduces taste function, particularly, to sour stimuli

Authors: *J. RIOS-PILIER, A. CLEMENTS, R. LUNDY, R. F. KRIMM
Anatom. Sci. and Neurobio., Univ. of Louisville, Louisville, KY

Abstract: BDNF-TrkB signaling regulates taste bud innervation, both during development and in adulthood. In the central nervous system, BDNF can function as a neuromodulator and influence neuronal function. In the taste bud, Car4+ cells, which transduce sour taste, express BDNF and synapse with TrkB expressing nerve fibers. However, it is unclear whether BDNF-TrkB signaling plays a functional role in taste. To test this possibility, we used a chemical-genetic approach, which allowed TrkB-signaling to be reversibly blocked with 1-NMPP1 in mice with a specific point mutation (*TrkB*^{F616A}). *TrkB*^{F616A} and wild type mice were injected with 1-NMPP1, three hours before taste function was assessed, using both chorda tympani (CT) whole nerve responses and brief-access behavioral tests. There was an overall decrease in taste responses to all stimuli tested. To determine if one stimulus was impacted more than others CT

responses were plotted relative to 0.1M NH_4Cl responses. Relative CT responses to 0.02M Citric acid and 0.01N HCl were significantly reduced in *TrkB*^{F616A} treated with 1-NMPP1 compared to wild type mice with 1-NMPP1 ($p<0.017$ and $p<0.008$, respectively) and *TrkB*^{F616A} vehicle treated mice ($p<0.036$ and $p< 0.033$, respectively). There were no behavioral taste differences in brief-access tests between *TrkB*^{F616A} and wild type mice when treated with 1-NMPP1. We also examined taste bud innervation in *TrkB*$^{CreER/+}$:tdtomato:*TrkB*^{F616A} mice after 3 hours of 1-NMPP1 to confirm that any changes in taste function were not a result of morphological changes in TrkB afferent fibers. There were no differences in branching patterns of TrkB expressing fibers after blocking TrkB signaling for three hours. These data suggests that BDNF-TrkB signaling may modulate taste function, especially on neurons innervating taste receptor cells transducing sour taste.

Disclosures: **J. Rios-Pilier:** None. **A. Clements:** None. **R. Lundy:** None. **R.F. Krimm:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.01/D16

Topic: B.02. Ligand-Gated Ion Channels

Support: FONDECYT (no. 1120156)

Basal Center of Excellence (PFB 12/2007)

Title: Role of Wnt-5a on GluN2B expression

Authors: *E. RAMOS[1,2], N. C. INESTROSA[1,2]
[1]Pontificia Univ. Católica, Santiago de Chile, Chile; [2]Ctr. for Aging and Regeneration, Santiago, Chile

Abstract: The Wnt-5a ligand is a secreted glycoprotein which interacts with the G-protein coupled receptor Frizzled 9. This interaction activates PLC and triggers the intracellular calcium increase through the non-canonical Wnt pathway. Among their roles, Wnt-5a is able to increase the clustering of PSD-95, also increases nitric oxide (NO) production, stimulates the translocation of NMDA receptor to the plasma membrane and up regulates NMDA currents. However, the specific Wnt-5a pathway involved in the regulation of the NMDAR function and the postsynaptic density structure is not clearly understood. Here we provide data showing that the activation of the Heme Regulated Eukaryotic Initiation Factor 2α (eIF2α) kinase (HRI), a kinase involved in memory consolidation, is essential to explain how Wnt-5a modulates NMDAR function through the production of NO. Using primary neuron cultures, synaptosomes from C57/BL/6J mouse, a specific inhibitor (iHRI) and genetic tools, we found that HRI kinase

has a role in GluN2B expression. Moreover, Wnt-5a treatment increases GluN2B expression generating new functional receptors, modulating postsynaptic density structure and dendritic spine number. All these effects are abolished with the iHRI. Our results suggest that NO produced by Wnt-5a, activates HRI kinase which in turn, phosphorylates eIF2α factor, allowing the translation of the GluN2B subunit of NMDAR to the postsynaptic density.

Disclosures: E. Ramos: None. **N.C. Inestrosa:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.02/D17

Topic: B.02. Ligand-Gated Ion Channels

Title: Postsynaptic serine racemase regulates NMDA receptors

Authors: *J. M. WONG[1,2], E. V. BARRAGAN[1,2], J. A. GRAY[1,3]
[1]Ctr. for Neurosci., [2]Neurosci. Grad. Group, [3]Dept. of Neurol., Univ. of California Davis, Davis, CA

Abstract: NMDA receptors (NMDARs) are glutamate-gated ion channels which uniquely require a co-agonist for activation: either glycine or D-serine. The identity of the co-agonist is developmentally regulated and spatially restricted in the brain, with many synapses using glycine early on and later switching to D-serine. Despite the importance of these co-agonists in regulating excitatory synaptic transmission, neuronal function, and synaptic plasticity, the source and regulation of D-serine remains controversial. Although D-serine and its biosynthetic enzyme serine racemase (SR) were originally thought to be localized in astrocytes, recent studies using SR constitutive & conditional knock-out mice combined with more selective antibodies instead suggest a predominantly neuronal localization. Furthermore, SR both complexes and colocalizes with PSD-95 suggesting that SR is localized at the postsynaptic density. Thus, to examine the potential role of postsynaptic SR in synaptic physiology, we assessed synaptic AMPAR- and NMDAR-EPSCs in SR conditional knockout (SRcKO) mice (generous gift of Dr. Joseph Coyle). We injected neonatal SRcKO mice with AAV expressing Cre-recombinase and GFP and obtained simultaneous dual whole-cell recordings from transduced and neighboring control neurons. This genetic approach utilizes sparse, localized transduction of single cells in order to rigorously analyze the cell-autonomous effects of SR deletion while avoiding presynaptic effects and the circuit-wide effects of global genetic or pharmacologic NMDAR manipulations. Synaptic recordings at Schaffer collateral-CA1 and perforant path-dentate gyrus (DG) synapses in the hippocampus from P45-60 mice show an approximately 40% reduction in NMDAR-EPSC amplitude in CA1 and a 70% reduction in DG in transduced versus control neurons while no effects of SR deletion were detectable in earlier development (P14-P21). AMPAR-EPSC

amplitudes were not significantly different from control at any assessed time point in either CA1 or DG. Overall, these results suggest a cell-autonomous neuronal regulation of NMDAR co-agonist occupancy by postsynaptic SR.

Disclosures: J.M. Wong: None. **E.V. Barragan:** None. **J.A. Gray:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.03/D18

Topic: B.02. Ligand-Gated Ion Channels

Support: RA-H is supported by an Omani Government Ministry of Higher Education Scholarship.

XC and WL were supported by a Beijing University Undergraduate Education grant.

Title: Adenosine A2A - D2 dopamine receptor modulation of NMDA responses in rat substantia nigra dopaminergic neurones

Authors: R. AL-HOSNI[1], F. CHERCHI[3], X. CAI[4], W. LEI[5], Z. HUANG[6], E. COPPI[3], *A. J. GIBB[2]
[1]Dept. of Neuroscience, Physiol. & Pharmacol., [2]Univ. Col. London, London, United Kingdom; [3]Dept. of Neuroscience, Psychology, Drug Res. and Child Hlth., Univ. of Florence, Scandicci, Italy; [4]Med., UCSD, La Jolla, CA; [5]Yale Univ., New Haven, CT; [6]Pharmacol., Beijing Univ., He Bei, China

Abstract: Dopamine receptor signalling is essential for normal basal ganglia function but in Parkinson's Disease (PD) substantia nigra (SNc) dopaminergic (DAergic) neurons degenerate with consequent loss of dopamine signalling. SNc DAergic neurons express D2 autoreceptors (D2Rs) and somatodendritically released dopamine mediates a negative feedback via D2Rs on DAergic neuron activity. D2Rs have been shown to mediate inhibition of NMDA responses in both hippocampus and striatum while $G_{\alpha s}$-coupled adenosine A2A receptors (A2ARs) have the potential to counteract the action of $G_{\alpha i}$ -coupled D2Rs. Here we tested whether D2R activation with ropinirole, a D2 receptor agonist currently used in PD therapy, modulates DAergic neuron NMDA responses in the SNc. To prevent D2R desensitization, a GRK 2/3 inhibitor, compound-101, was introduced via the intracellular pipette solution. Lastly, an A2AR agonist, CGS-21680, was applied in the presence and absence of ropinirole to test for any A2AR - D2R interaction. Whole-cell patch clamp recordings were made from DAergic neurons in the SNc of acute midbrain slices from neonatal (P7, P21 and P28) rats. DAergic neurons were identified by the presence of a prominent hyperpolarisation-activated inward current (In P7 rats, amplitude, 173 ±

34 pA; activation time constant, 797 ± 113 ms; mean ± SE, n = 11) in response to a voltage step from -60 to -120 mV. In each cell, two successive responses to 20 υM NMDA with 10 uM glycine were recorded. In P7 rats, control experiments gave NMDA responses of 919 ± 89 pA (1^{st} response) and 881 ± 113 pA, n = 11 (2^{nd} response) while in P28 rats the control NMDA responses were 790 ± 110 pA (1^{st} response) and 800 ± 93 pA, n = 10 respectively. In P7, P21 and P28 rats, upon application of 200 nM ropinirole, the steady state NMDA current was not significantly changed suggesting D2R activation may not modulate NMDARs in neonatal rat SNc. It is also possible a D2 effect might be observed in older rats. Furthermore, limitations involved in the whole-cell patch-clamp configuration, such as dilution of cellular components could affect the result. An A2AR agonist, CGS-21680 (1 or 10 υM) was applied to P28 DAergic neurons to determine the effect of activating this $G_{\alpha s}$ - coupled receptor which may be in a D2R-A2AR heteromeric complex. 1 υM CGS-21680 gave a 46 ± 16 % increase in inward current (P = 0.06, paired t-test; n = 12) while in the presence of 10 υM CGS-21680 and 200 nM ropinirole, the NMDA current increased 59 ± 16 % (P = 0.002, n = 16).

Disclosures: **R. Al-Hosni:** None. **F. Cherchi:** None. **X. Cai:** None. **W. Lei:** None. **Z. Huang:** None. **E. Coppi:** None. **A.J. Gibb:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.04/D19

Topic: B.02. Ligand-Gated Ion Channels

Support: Sponsored Research Agreement from Sage Therapeutics

Title: Positive allosteric modulation as a potential therapeutic strategy in anti-NMDA receptor encephalitis

Authors: N. WARIKOO[1], S. BRUNWASSER[1], A. BENZ[1], J. J. DOHERTY[4], M. C. LEWIS[4], M. QUIRK[4], S. M. PAUL[4], L. PICCIO[2], C. F. ZORUMSKI[1], G. DAY[2], *S. J. MENNERICK[3]
[1]Psychiatry, [2]Neurol., Washington Univ. Sch. of Med., St. Louis, MO; [3]Psychiatry, Washington Univ. Sch. of Med., Saint Louis, MO; [4]Sage Therapeut., Cambridge, MA

Abstract: NMDA receptors (NMDARs) are ionotropic glutamate receptors important for synaptic plasticity, memory, and neuropsychiatric health. NMDAR hypofunction contributes to multiple disorders, including anti-NMDAR encephalitis (NMDARE), a neuro-autoimmunological condition characterized by autoantibodies against CNS NMDARs and by NMDAR internalization. Symptoms include psychosis, seizures, cognitive deficits, and death. With immunotherapy, about 75% of patients recover. However, there remains unmet therapeutic need; immunosuppression onset can be slow, and there is incomplete recovery and relapse. Here

we characterize functional/pharmacological consequences of antibody exposure and introduce an alternative/adjunct treatment using positive allosteric modulators (PAMs) of NMDAR function. Staining of fixed mouse brain tissue slices incubated in cerebrospinal fluid (CSF) from a NMDARE patient yielded labeling with anti-human secondary antibody yielded labeling consistent with NMDAR distribution, confirming clinical diagnosis. Incubation (48 h) of rat hippocampal neurons in NMDARE patient CSF, but not asymptomatic CSF or multiple sclerosis CSF, exhibited attenuated NMDA-induced current. We characterized residual NMDAR function. Using open-channel blocker kinetics, we detected no change in P_{open} of remaining surface NMDARs. There was also no preferential loss of synaptic versus extrasynaptic or of GluN2A versus GluN2B NMDARs. These experiments suggest that broad-spectrum PAMs could be a viable intervention. We employed two PAMs, SGE-301 and SGE-550, that are analogues of the natural cholesterol metabolite 24S-hydroxycholesterol. Co-application of SGE-301 or SGE-550 with CSF for 24 h, following an initial 24 h in CSF alone, restored NMDAR function, even in the nominal absence of PAMs during evaluation. Memantine kinetics following 24 h SGE-301/550 incubation revealed increased P_{open}, consistent with PAM action. Furthermore, persisting restorative effects of SGE-301/550 were reversed by brief incubation with γ-cyclodextrin, a cyclic oligosaccharide used as a steroid/sterol scavenger. Thus, it appears that the effects of SGE-301/550 lie in persisting drug presence. Together, our studies demonstrate restoration of NMDAR functionality through augmentation of remaining NMDAR function. Further studies of the pathogenesis of NMDARE and PAM intervention will inform new treatments for this disorder and others involving NMDAR hypofunction.

Disclosures: **N. Warikoo:** None. **S. Brunwasser:** None. **A. Benz:** None. **J.J. Doherty:** A. Employment/Salary (full or part-time):; Sage Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Sage Therapeutics. **M.C. Lewis:** A. Employment/Salary (full or part-time):; Sage Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Sage Therapeutics. **M. Quirk:** A. Employment/Salary (full or part-time):; Sage Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Sage Therapeutics. **S.M. Paul:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Sage Therapeutics. **L. Piccio:** None. **C.F. Zorumski:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Sage Therapeutics. **G. Day:** None. **S.J. Mennerick:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Sage Therapeutics.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.05/D20

Topic: B.02. Ligand-Gated Ion Channels

Title: Effects of amines on GluN1/GluN2A and GluN1/GluN2B subtypes of NMDA receptor

Authors: *Y. YAMADA, T. YABUKI, K. NISHII
Fac. of Engineering, Kindai.Univ., Higashihiroshima-Shi, Japan

Abstract: NMDA-type glutamate receptors (NMDA receptors) are widely distributed in the central nervous system and play critical roles in synaptic plasticity and excitotoxicity. Dysfunctions of NMDA receptors are involved in several central nervous system disorders, including stroke, chronic pain and schizophrenia. There are a few NMDA receptor antagonists available for clinical use, including ketamine and most importantly, mematine, both of which act as channel blockers. Polyamines, such as putrescine, spermine and spermidine, are poly basic aliphatic amines that were ubiquitously present in prokaryotic and eukaryotic cells. Extracellular polyamines have multiple effects on NMDA receptors. Spermine is known to inhibit or activate the channel activity of NMDA receptor subunits dependent manner. Only NMDA receptors containing the GluN2B subunits display polyamine potentiation. The study of the channel block activity with the polyamine derivative is applied to a study of the structure activity correlation of the receptor, development of the brain function or improvement medicine. It was known that over 1000 different ingredients such as sugars, amino acids, amines and organic acids are included in sake, traditional Japanese alcoholic beverage made from rice. The components of sake were measured using CE-TOFMS and 186 components were identified. Twelve amines of 186 components were examined the effects on the channel activity of GluN1/GluN2A- and GluN1/GluN2B- subtypes of NMDA receptor using *Xenopus* oocyte expression system and elevated plus-maze test. Agmatine, putresine, tyramine, 2-phenylethylamine, ornithine, choline, 5-oxoproline, cysteic acid,and N-acetylglutamic acid inhibited the activity of both receptors. It was indicated that isoamylamine and betaine acts as antagonists of the GluN1/GluN2B receptor, but activators for GluN1/GluN2A receptor. Trimethylamine N-oxide promoted the activity of both receptors. Isoamylamine, 2-phenylethylamine and ornithine indicated significant anxiolytic effects by an elevated plus-maze test. Observed differences in activity between the receptor subtypes, it may be able to elucidate the mechanism of amines action on the NMDA receptor channel opening. These results indicate that these compounds from sake will be useful for the elucidation of regulation of NMDA receptors and the development of agents that target and alter NMDA receptor function may have therapeutic benefit.

Disclosures: **Y. Yamada:** None. **T. Yabuki:** None. **K. Nishii:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.06/DP02/D21 (Dynamic Poster)

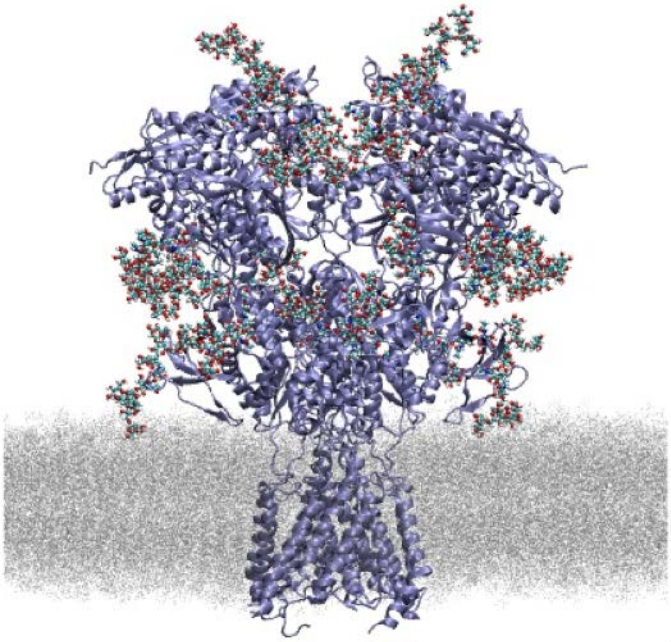
Topic: B.02. Ligand-Gated Ion Channels

Support: Sherlock cluster at the Stanford Research Computing Center

Title: Computer simulations of NMDA receptors with all-atom resolution: A new tool to study the dynamics of NMDARs

Authors: *A. SINITSKIY, V. S. PANDE
Chem., Stanford Univ., Stanford, CA

Abstract: N-methyl-D-aspartate (NMDA) receptors are transmembrane proteins playing the key role in various cognitive processes, including learning and memory formation. Understanding the structure and dynamics of NMDA receptors (NMDARs) with atomic resolution is critically important for knowing how the brain works, and for developing new drugs to treat NMDAR-associated disorders. Several full all-atom structures of NMDARs in closed and inhibited functional states have recently been determined by X-ray crystallography and cryoEM. However, the dynamics of NMDARs at atomic resolution, especially on submillisecond timescales, is not sufficiently understood. It is also unclear how the published atomic structures of NMDARs relate to various functional states of NMDARs established from electrophysiological experiments. Here, we report the results of our computer simulations of an apo and an agonist-and-coagonist-bound NMDARs. To the best of our knowledge, these are the first reported all-atom simulations of a full NMDAR under physiological conditions. In total, 300 molecular dynamics (MD) trajectories were generated, each 100 ns long, leading to the aggregate sampling of 30 μs. Markov state models (MSMs) were used to estimate the equilibrium and nanosecond-to-microsecond dynamic properties of NMDARs from these finite-length trajectories.
Our simulations have revealed that the conformational transitions in the amino terminal domain (ATD) of NMDARs, previously discovered by cryoEM, occur much faster (~ 0.4 μs) than transitions between various functional states of the receptor (~ 1-100 ms). For this reason, we conclude that despite significant structural differences between four 'non-active' cryoEM structures of NMDARs (up to 10-20 Å), these four structures (PDB codes 5FXH, 5FXI, 5FXJ, 5FXK) may refer to the same functional state of the receptor, namely one of the closed states. Overall, computational modeling, and specifically all-atom MD simulations, can serve as a powerful tool for investigating NMDARs and yield information unavailable from experimental techniques alone.



Disclosures: **A. Sinitskiy:** None. **V.S. Pande:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Andreessen Horowitz. F. Consulting Fees (e.g., advisory boards); Schrodinger, LLC, Globavir, Apeel Inc, Freenome Inc, Omada Health, Patient Ping, Rigetti Computing.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.07/D22

Topic: B.02. Ligand-Gated Ion Channels

Support: NIH R00AG041225

NIH T32GM08061

Title: Regulation of NMDAR trafficking by protein phosphatase 1

Authors: *A. M. CHIU, L. W. BARSE, A. SANZ-CLEMENTE
Northwestern Univ., Chicago, IL

Abstract: N-methyl-D-aspartate receptors (NMDARs) are glutamate receptors that are responsible for the molecular basis of learning and memory through their ability to control processes such as synaptic plasticity and synaptic maturation. NMDARs are able to achieve these phenomena through their ability to initiate specific intracellular cascades in response to receptor

activation. The subunit composition and localization of the receptor regulate which intracellular cascades are triggered upon NMDAR activation. Thus, a key aspect of NMDAR regulation and function is tight control over trafficking of the receptor.
Posttranslational modifications are a crucial component of receptor trafficking. We have previously investigated how phosphorylation within the PDZ-binding domain of the GluN2B subunit of NMDARs results in the disruption of NMDARs from scaffolding proteins to reduce the synaptic content of the receptor. More recently, we sought to understand how the complementary reaction, dephosphorylation of this site, is controlled. Using primary cortical neurons and acute cortical slices, we have identified that calcium influx into the cell following activation of extrasynaptic GluN2B-containing NMDARs triggers the dephosphorylation of surface-expressed GluN2B-containing NMDARs.
In an effort to better understand the mediators of this dephosphorylation, we screened protein phosphatases and have identified protein phosphatase 1 (PP1) as being responsible for dephosphorylating the GluN2B PDZ-binding domain. However, because PP1 is a constitutively active phosphatase, there exist a number of molecular modes of regulation to control its activity. These include inhibitory PP1 phosphorylation, association with endogenous inhibitor proteins, and subcellular substrate targeting. Because canonical PP1 regulation involves a mixture of these forms of regulation, we are examining how each of these mechanisms may play a role in regulating PP1 activity against GluN2B's PDZ-binding domain. Ultimately, we believe that understanding how PP1 regulates this posttranslational modification and its consequence on NMDAR trafficking and localization will help to understand how physiological NMDAR trafficking occurs and how this is altered in disease.

Disclosures: **A.M. Chiu:** None. **L.W. Barse:** None. **A. Sanz-Clemente:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.08/D23

Topic: B.02. Ligand-Gated Ion Channels

Support: NIH Grant GM120844

Title: Bortezomib induces neuropathic pain through PKC-mediated activation of presynaptic nmda receptors in spinal cords

Authors: *H.-L. PAN[1], S.-R. CHEN[2]
[1]Univ. Texas-MD Anderson Cancer Ctr., Houston, TX; [2]Anesthesiol. & Perioperative Med., Univ. of Texas MD Anderson Cancer Ctr., Houston, TX

Abstract: The glutamate N-methyl-D-aspartate receptors (NMDARs) at the spinal cord level are critically involved in the synaptic plasticity associated with neuropathic pain. In this study, we determined whether treatment with bortezomib, a proteasome inhibitor, affects the NMDAR activity of spinal dorsal horn neurons. Systemic treatment with bortezomib in rats did not significantly affect postsynaptic NMDAR currents elicited by puff application NMDA directly to dorsal horn neurons. Bortezomib treatment markedly increased the baseline frequency of miniature excitatory postsynaptic currents (EPSCs), which was completely normalized by the NMDAR antagonist 2-amino-5-phosphonopentanoic acid (AP5). AP5 also reduced the amplitude of monosynaptic EPSCs evoked by dorsal root stimulation in bortezomib-treated, but not vehicle-treated, rats. Furthermore, inhibition of protein kinase C (PKC) with chelerythrine fully reversed the increased frequency of miniature EPSCs and the amplitude of evoked EPSCs in bortezomib-treated rats. Intrathecal injection of AP5 and chelerythrine both profoundly attenuated mechanical allodynia and hyperalgesia induced by systemic treatment with bortezomib. In addition, treatment with bortezomib induced striking membrane translocation of PKC-βII, PKC-δ, and PKC-ε in the dorsal root ganglion. Our findings indicate that bortezomib treatment potentiates nociceptive input from primary afferent nerves via PKC-mediated tonic activation of presynaptic NMDARs. Targeting presynaptic NMDARs and PKC at the spinal cord level may be an effective strategy for treating chemotherapy-induced neuropathic pain.

Disclosures: H. Pan: None. **S. Chen:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.09/D24

Topic: B.02. Ligand-Gated Ion Channels

Support: R01 MH045817

F31 MH105056

T32 NS073548

T32 NS007433

Title: Memantine inhibition of native synaptic NMDA receptors is Ca^{2+}-dependent

Authors: *N. V. POVYSHEVA[1], N. G. GLASGOW[2], J. W. JOHNSON[1]
[1]Neurosci., [2]Neurobio., Univ. of Pittsburgh Dept. of Neurosci., Pittsburgh, PA

Abstract: Memantine is an NMDA receptor (NMDAR) channel blocker approved for treatment of Alzheimer's disease, and with broad potential as a neuroprotective agent. Several studies have

suggested that memantine inhibits extrasynaptic NMDARs, which are hypothesized to be strongly linked to excitotoxic pathways, more effectively than synaptic NMDARs. Our investigation of mechanisms that could be responsible for preferential inhibition of extrasynaptic NMDARs revealed that memantine stabilizes a Ca^{2+}-dependent desensitized state of GluN1/2A NMDARs. Here we address whether this novel mechanism of action of memantine is applicable to native neocortical NMDARs. We studied effects of memantine on NMDAR excitatory postsynaptic currents (EPSCs) in layer II/III pyramidal neurons from prefrontal cortical slices of wild-type mixed background C57BL/6J, BALB/cJ mice of either sex. NMDAR EPSCs were evoked by extracellular stimulation at the border of layer VI and white matter. Memantine (10 μM) was applied in two conditions: (1) "High Ca^{2+}" condition, with 2 mM external Ca^{2+} and no Ca^{2+} buffer in the internal solution; and (2) "Low Ca^{2+}" condition, with 1 mM external Ca^{2+} and 10 mM BAPTA in the internal solution. External solution also contained 0.5 mM Mg^{2+} and 10 μM glycine. NMDAR EPSCs were isolated by addition of GABAA, AMPA and kainate receptor blockers. Since Ca^{2+}-dependent desensitization is more pronounced toward the end of stimulus trains, we assessed effects of memantine on the amplitude of the 10^{th} response to trains of 10 stimuli. Strikingly, whereas under high Ca^{2+} conditions 10 μM memantine decreased the amplitude of the 10^{th} response by 49 ± 7%, under low Ca^{2+} conditions the decrease was only 22 ± 2% ($p<0.01$). Our results support the conclusion that memantine stabilizes Ca^{2+}-dependent desensitized states of native as well as recombinant NMDARs. However, interpretation of these experiments could be complicated by the presence of presynaptic NMDARs at the stimulated synapses. Memantine block of presynaptic NMDARs could affect Ca^{2+} influx more substantially in high than in low external Ca^{2+}, resulting in a more pronounced reduction of EPSC amplitude under high Ca^{2+} conditions. Thus, we assessed potential presynaptic effects of memantine by measuring the paired- pulse ratio (PPR) before and after memantine application. Under both high and low Ca^{2+} conditions, the PPR was not significantly affected by memantine. Elucidating memantine mechanisms of action can provide important information for development of more effective neuroprotective drugs. Here we identify NMDAR Ca^{2+}- dependent desensitization as a novel target for drug development.

Disclosures: **N.V. Povysheva:** None. **N.G. Glasgow:** None. **J.W. Johnson:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.10/D25

Topic: B.02. Ligand-Gated Ion Channels

Support: Charles University Grant Agency (GAUK) no. 880216.

Czech Science Foundation (GACR): P304/12/G069 and 17-02300S

Technology Agency of the Czech Republic: TE01020028

Title: Molecular insight into the N-methyl-D-aspartate receptor channel gating

Authors: *M. LADISLAV[1,2], J. CERNY[3], J. KRUSEK[1], K. SKRENKOVA[1], M. HORAK[1], A. BALIK[2,1], L. VYKLICKY JR[1,2]
[1]Cell. Neurophysiol., Inst. of Physiol. CAS, Prague, Czech Republic; [2]Cell. Neurophysiol., Inst. of Physiol. CAS, Vestec, Czech Republic; [3]Structural Bioinformatics of Proteins, Inst. of Biotech. CAS, Vestec, Czech Republic

Abstract: N-methyl-D-aspartate receptors (NMDARs) are heterotetramers containing two obligatory glycine-binding (GluN1) and two glutamate/glycine-binding (GluN2/3) subunits. These receptors mediate excitatory synaptic transmission in the central nervous system and it has been shown that dysregulation of NMDARs is involved in the pathophysiology of neurological and psychiatric disorders. Channel opening is the key step in the NMDAR gating that allows the flux of ions including Ca^{2+} across the membrane. Several lines of evidence indicate that the rearrangement of M3 helices in activated receptor makes the central cavity of the channel accessible therefore implying a crucial role of the M3-S2 linkers in channel opening. To answer the fundamental question, what are the initial steps in NMDAR channel opening, we embarked on functional, molecular biology and molecular dynamics studies of GluN1/GluN2B receptors and focused on the M3 and initial segments of the M3-S2 linkers. The results of our experiments show that deletion mutations and glycine substitution mutations in the M3-S2 linker of GluN1 and GluN2B subunits profoundly affect the NMDAR channel function: i. Mutated NMDARs open spontaneously and as a consequence of receptor activation by a single agonist; ii. The effect of deletions is stratified - spontaneous activity and single-ligand induced responses are more pronounced at deletions closer to the M3 helix; iii. The degree of spontaneous activity and single-agonist responses, as well as the size of the region affected by deletion mutations, differ for GluN1 and GluN2B subunits; iv. Irrespective of whether deletions were introduced in GluN1 or GluN2B subunits, the application of glutamate or glycine promoted receptor channel activity; v. Irrespective of whether the deletion was introduced to the M3-S2 linker of GluN1 and GluN2B, responses induced by glycine were (on average) larger than those induced by glutamate; vi. None of the single- and double-deletions in the linker region mimicked receptor activation by a single-agonist (insensitive to glutamate and sensitive to glycine or vice versa insensitive to glycine and sensitive to glutamate); vii. There is a bell-shaped relation between the spontaneous activity and single-agonist induced activity. Combining functional data with those of results of computational biology we show that the extracellular channel gate is formed by GluN1(L657) and GluN2B(I655) (LILI motif). Our data provide new insight into the mechanism of NMDAR ion channel gating and describes LILI motif crucial for the transduction of the energetics of agonist binding to the ligand binding domain to pore opening.

Disclosures: **M. Ladislav:** None. **J. Cerny:** None. **J. Krusek:** None. **K. Skrenkova:** None. **M. Horak:** None. **A. Balik:** None. **L. Vyklicky Jr:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.11/D26

Topic: B.02. Ligand-Gated Ion Channels

Support: Russian Science Foundation grant 16-15-10192

European Union Seventh Framework Program FP7/2007–2013 under the project DESIRE grant agreement 602531

"Fédération pour la Recherche sur le Cerveau" (FRC) grant R15018AA

French "Institut National de la Santé et de la Recherche Médicale" (INSERM)

Academy of Finland grant 277442

Program for Competitive Growth at Kazan Federal University

Title: Epilepsy-related mutations of GluN2A subunit of NMDA receptors

Authors: *D. A. SIBAROV[1], N. BRUNEAU[2], S. M. ANTONOV[1], P. SZEPETOWSKI[2], N. BURNASHEV[2], R. GINIATULLIN[3]
[1]Comparative Neurosci., IEPHB RAS, Sankt-Peterburg, Russian Federation; [2]INMED, Aix-Marseille Univ., Marseille, France; [3]Univ. of Eastern Finland, Kuopio, Finland

Abstract: Genetic variants of the glutamate activated NMDA receptor (NMDAR) subunit GluN2A are associated with the hyperexcitable states manifested by epileptic seizures and interictal discharges in patients with disorders of the epilepsy-aphasia spectrum. Deciphering the consequences of pathogenic GRIN2A variants would surely help in better understanding of the underlying mechanisms. This emphasizes the need for functional studies to unravel the basic functional properties of each specific NMDAR variant. In the present study, we have used patch-clamp recordings to evaluate kinetic changes of mutant NMDARs reconstituted after co-transfection of cultured cells with the appropriate expression vectors. Three previously identified missense variants found in patients or families with disorders of the epilepsy-aphasia spectrum and situated in the N-terminal domain (p.Ile184Ser) or in the ligand-binding domain (p.Arg518His and p.Ala716Thr) of GluN2A were studied in both the homozygous and heterozygous conditions. Relative surface expression and current amplitude were significantly reduced for NMDARs composed of mutant p.Ile184Ser and p.Arg518His, but not p.Ala716His, as compared with WT NMDARs. Amplitude of whole-cell currents was still drastically decreased when WT and mutant p.Arg518His-GluN2A subunits were co-expressed, suggesting a dominant-negative mechanism. Activation times were significantly decreased in both

homozygous and heterozygous conditions for the two p.Ile184Ser and p.Arg518His variants, but not for p.Ala716His. Deactivation also significantly increased for p.Ile184Ser variant in the homozygous but not the heterozygous state while it was increased for p.Arg518His in both states. Our data indicate that p.Ile184Ser and p.Arg518His GluN2A variants both impacted on NMDAR function, albeit differently, whereas p.Ala716His did not significantly influence NMDAR kinetics, hence partly questioning its direct and strong pathogenic role. This study brings new insights into the functional impact that GRIN2A variants might have on NMDAR kinetics, and provides a mechanistic explanation for the neurological manifestations seen in the corresponding human spectrum of disorders.

Disclosures: D.A. Sibarov: None. **N. Bruneau:** None. **S.M. Antonov:** None. **P. Szepetowski:** None. **N. Burnashev:** None. **R. Giniatullin:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.12/D27

Topic: B.02. Ligand-Gated Ion Channels

Title: The A, B, C and D of NMDA receptor modulation

Authors: C. T. BOT[1], *A. R. OBERGRUSSBERGER[2], I. RINKE-WEIß[2], S. STÖLZLE-FEIX[2], N. BECKER[2], E. DRAGICEVIC[2], C. HAARMANN[2], A. BRÜGGEMANN[2], M. GEORGE[2], N. FERTIG[2]
[1]Nanion Technologies Inc, Livingston, NJ; [2]Nanion Technologies GmbH, Muenchen, Germany

Abstract: N-Methyl-D-aspartate (NMDA) receptors are a member of the ionotropic glutamate receptor family of ligand-gated ion channels that mediate the majority of excitatory neurotransmission in the mammalian CNS. They are expressed primarily in the CNS but also in peripheral locations such as pancreatic islet cells, sensory nerve terminals in skin and cardiac ganglia. Seven subunits of the NMDA receptor have been identified, NR1, NR2A-D and NR3A-B2, they assemble as a tetramer consisting of two NR1 subunits and either two NR2 subunits or a combination of NR2 and NR3 subunits. Activation of NMDA receptors requires the simultaneous binding of glutamate and glycine. Calcium entry through NMDA receptors plays an important role in development and synaptic plasticity and is proposed to underlie higher processes such as learning and memory. It is also proposed to play a role in a number of neurological diseases such as epilepsy and Alzheimer's. Indeed, memantine is an NMDA antagonist which has been approved for the treatment of moderate to severe Alzheimer's. NMDA antagonists may also be targets for the treatment of neuropathic pain, major depression and Parkinson's disease. We present data of NMDA receptor combinations NR1 with either NR2A, 2B, 2C or 2D expressed in HEK cells recorded on a high throughput automated patch

clamp system. NMDA-mediated responses were elicited using glutamate and glycine. Mean current amplitudes varied from -127 ± 9 pA for NR1/NR2C (n = 188), -288 ± 21pA for NR1/NR2D (n = 191), -1.72 ± 0.19 nA for NR1/NR2A (n = 360) and -2.99 ± 0.33 nA for NR1/NR2B (n = 372) receptor combinations. The currents mediated by NR1/NR2A were potentiated by pregnenolone sulfate with an EC_{50} of approximately 40 µM (n = 284). NR1/NR2B-mediated currents were potentiated by spermine with an EC_{50} of 134 µM (n = 290) and blocked by ketamine with an IC_{50} of 2.26 ± 0.38 µM (n = 321). The currents mediated by NR1/NR2C and NR1/NR2D were potentiated by CIQ with an EC_{50} of 4.3 µM (n = 169) and 5.5 µM (n = 149), respectively. Interestingly, we could detect differences in potency of compounds such as ifenprodil on NR1/NR2A versus NR1/NR2B containing receptors. In addition to NMDA expressed in cell lines, we have also investigated the presence of NMDA receptors in stem cell-derived neurons. Glutamate (in combination with glycine) was used to elicit NMDA-mediated responses using automated patch clamp and MEA. In this way, we could record whole cell patch clamp data and compare this with extracellular field potential recordings in intact neuronal networks of stem cell-derived neurons.

Disclosures: C.T. Bot: A. Employment/Salary (full or part-time):; Nanion Technologies Inc. **A.R. Obergrussberger:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH. **I. Rinke-Weiß:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH. **S. Stölzle-Feix:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH. **N. Becker:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH. **E. Dragicevic:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH. **C. Haarmann:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH. **A. Brüggemann:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH. **M. George:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH. **N. Fertig:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.13/D28

Topic: B.02. Ligand-Gated Ion Channels

Support: NIH R01N5052669

Title: Ca^{2+}/calmodulin mediates strong negative coupling of NMDA receptor gating

Authors: *G. IACOBUCCI[1], G. K. POPESCU[2]
[1]Biochem., State Univ. of New York At Buffalo, Buffalo, NY; [2]Biochem., Univ. at Buffalo, SUNY, Buffalo, NY

Abstract: In the central nervous system, NMDA receptors generate large and highly regulated Ca^{2+} signals, which are critical for synaptic development, plasticity, and apoptosis. NMDA receptors are highly clustered at postsynaptic dendritic spines and exhibit spatially heterogeneous surface expression along dendrite arbors. Whether the spatial arrangement of NMDA receptors affects their signal is unknown. Synaptic NMDA receptor currents are subject to Ca^{2+}-dependent inactivation (CDI), a type of activity-dependent inhibition that depends on intracellular Ca^{2+} and calmodulin (CaM). We asked whether Ca^{2+} influx through a single NMDA receptor influences the activity of nearby NMDA receptors. We used cell-attached current recordings from GluN1-2a/GluN2A receptors expressed in HEK293 cells and from NMDA receptors native to dissociated hippocampal neurons. We measured transition probabilities (P_O) in multi-channel patches and analyzed these data with a coupled Markov chain model of cooperative gating to calculate the coupling factor, κ. In the absence of extracellular Ca^{2+}, we observed no cooperativity ($\kappa < 0.1$), whereas in 2 mM of external Ca^{2+}, both recombinant and native channels showed modest but significant cooperativity ($\kappa = 0.2$). This cooperativity was strongly reduced by pretreating cells with BAPTA-AM and by overexpressing a Ca^{2+}-insensitive CaM mutant. These results strongly indicate that NMDA receptor gating is coupled and the decrease in P_O depends on intracellular Ca^{2+} and CaM, as expected for CDI. Further, we altered NMDA receptor clustering by overexpressing PSD-95, a postsynaptic scaffolding protein and observed larger fluorescent puncta by immunofluorescence and much stronger cooperativity ($\kappa = 0.6$). Together, these results demonstrate Ca/CaM dependent negative cooperativity of NMDA receptor gating and suggest a possible role for channel clustering in activity-dependent NMDA receptor modulation.

Disclosures: G. Iacobucci: None. **G.K. Popescu:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.14/D29

Topic: B.02. Ligand-Gated Ion Channels

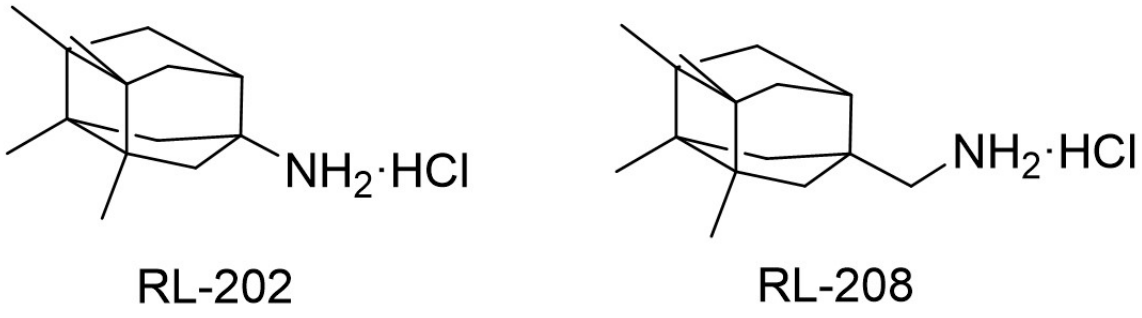
Support: NIH Grant MH045817

Ministerio do Ciencia e Innovación Project CTQ2011-22433

Title: Characteristics of NMDA receptor inhibition by the novel channel blockers RL-202 and RL-208

Authors: *M. B. PHILLIPS[1], R. LEIVA[2], S. VÁZQUEZ[2], J. W. JOHNSON[1]
[1]Neurosci., Univ. of Pittsburgh, Pittsburgh, PA; [2]Dept. of Pharmacology, Toxicology, and Therapeut. Chem., Univ. of Barcelona and IBUB, Barcelona, Spain

Abstract: NMDA receptors (NMDARs) are a class of ionotropic glutamate receptors (iGluRs) expressed at nearly all vertebrate synapses. NMDARs display a variety of properties unique amongst iGluRs, including dependence upon co-agonists, voltage-dependent Mg^{2+} block, slow kinetics, and permeability to Ca^{2+}. NMDAR activity is critical for many types of synaptic plasticity and is a key player in memory formation and learning. Conversely, aberrant NMDAR activation is implicated in a variety of nervous system disorders. Pharmacological targeting of NMDARs with channel blockers has shown therapeutic promise for protection from excitotoxicity as well as the treatment of Alzheimer's disease and major depressive disorder. Despite sharing similarities in binding site and mechanism of inhibition, the clinical utility of NMDAR channel blockers with differing structure can vary dramatically. Further investigation into how channel blockers differentially affect receptor function may provide insight into their varying clinical efficacy and aid in future drug design. Here we describe the synthesis and pharmacological characteristics of two novel NMDAR channel blockers, the polycyclic amines RL-202 and RL-208 (Figure 1). RL-202 and RL-208 were synthesized from 2,3-dimethylbutadiene and dimethyl acetylenedicarboxylate through a straightforward synthetic route. Recordings from NMDARs expressed in tsA201 cells revealed that both compounds share similarities with the well-characterized NMDAR channel blocker memantine, including moderate IC_{50} (RL-202 IC_{50} = 2.8 μM; RL-208 IC_{50} = 1.0 μM), strong voltage dependence of block, and second site inhibition. Second site inhibition is a form of antagonism exhibited by some, but not all, NMDAR channel blockers that involves drug binding to a site outside the NMDAR channel in the absence of agonist. Interestingly, both compounds display notably greater second site inhibition than memantine. Our findings suggest that these compounds could be useful tools for elucidating and differentiating mechanisms of NMDAR inhibition by channel blockers.



Structures of RL-202 and RL-208.

Disclosures: **M.B. Phillips:** None. **R. Leiva:** None. **S. Vázquez:** None. **J.W. Johnson:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.15/D30

Topic: B.02. Ligand-Gated Ion Channels

Title: PSD<->95 deficiency alters glutamatergic transmission in the prefrontal cortex during development

Authors: *A. A. COLEY, W.-J. GAO
Dept. of Neurobio. and Anat., Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: Postsynaptic density protein-95 (PSD-95) is a major regulator in the maturation of excitatory synapses by interacting and trafficking N-methyl-D-aspartic acid receptors (NMDAR's) and α-amino-3-hydroxy-5-methyl-4-isox-azoleproprionic acid receptors (AMPAR's) to the postsynaptic membrane of the dendritic spine. PSD-95 disruption has recently been connected to neuropsychiatric disorders such as schizophrenia and autism. However, the effects of PSD-95 deficiency within the prefrontal cortex (PFC)—a brain region responsible for cognition, working memory, emotional control, and sociability—has yet to be investigated. We hypothesize that PSD-95 deficiency will disrupt synaptic maturation during critical periods of neurodevelopment due to an increase in NMDAR/AMPAR-glutamatergic transmission that leads to impairments in PFC development and function. Therefore, using a PSD-95 knockout mouse model (PSD-$95^{-/-}$), we examined how PSD-95 deficiency affects NMDAR and AMPAR expression and function in the medial prefrontal cortex (mPFC) at postnatal days 21, 35 and 70, i.e., juvenile, adolescent and adult periods, respectively. We found significant increases in total protein levels of NMDAR subunits NR1, and NR2B, accompanied with a trending decrease in AMPAR subunit GluR1 during adolescence. Whole-cell patch clamp recordings of NMDA- and AMPA-mediated currents reveal significant increases in NMDAR/AMPAR-mediated current amplitude, indicating an increase in silent synapses. However, no significant changes in NMDAR/AMPAR protein expression or function were observed during juvenile time points, indicating that the adolescence age range is a critical period at which PSD-95 influences NMDAR and AMPAR expression levels in the mPFC. Behaviorally, using a 3-chamber social interaction task, we show PSD-$95^{-/-}$ mice show a lack of social exploration and novelty that could be indicative of social withdrawal symptoms. These data indicate that PSD-95 deficiency disrupts mPFC synaptic function and behavior at a critical age range. This study describes the importance of PSD-95 during neurodevelopment in the mPFC and its potential association in the pathogenesis of cognitive and learning deficits in schizophrenia and/or autism.

Disclosures: A.A. Coley: None. **W. Gao:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.16/D31

Topic: B.02. Ligand-Gated Ion Channels

Title: Chicken embryo nerve cells culture from forebrain is a useful *In vitro* model to screen compounds aimed at glutamate receptor ion channels

Authors: *M. F. FJELLDAL[1,3,4], J. E. JAKOBSSON[2,5], P. RISS[2,5], A. RING[3], I. SYLTE[7], L. M. EVENSETH[8], R. E. PAULSEN[1,6]
[1]Sch. of Pharm., [2]Dept. of Chem., Univ. of Oslo, Oslo, Norway; [3]Norwegian Def. Res. Establishment, Kjeller, Norway; [5]Dept. of Chem., [6]Sch. of Pharm., [4]Realomics Strategic Res. Initiative, Oslo, Norway; [7]Dept. of Med. Biol., [8]Mol. Pharmacol. and Toxicology, UiT - The Arctic Univ. of Norway, Tromsø, Norway

Abstract: The NMDA receptor ion channel is activated by the excitatory neurotransmitter glutamate and is present in virtually every nerve cell in the CNS. It is essential to neurophysiological processes and is implicated in pathological neurodegeneration after stroke, epilepsy, Alzheimer's, Parkinson's and other CNS diseases. The composition and localization of NMDAR subunits is dynamic and varies across the CNS during development. Rodent nerve cell cultures are widely used models to characterize drugs binding to the NMDA receptor. We have used cultures of embryonic chicken forebrain neurons as an alternative to mammalian cultures in order to screen for neuro-active compounds aimed at NMDA receptor subtype NR2B. We hypothesized that chicken embryo forebrain neurons, with an abundance of cortical cells, express NR2B-containing NMDA receptors. The predicted amino acid sequences of chicken NMDA receptor subunits are very similar to those of rat. Advantages associated with the chicken model are; a) cells are dissected from 10 day old embryos (from eggs, E10) and such embryos are not yet subject to EU regulations for animal experiments, b) the model is economical and does not require animal house facilities, c) there are less allergies associated with dissection or handling of chicken embryos and d) as they are easily accessible, embryos may be injected with drugs prior to dissection to test for neurotoxic or developmental effects (Bjørnstad et al., JPET 355: 386-96, 2015). We have measured NMDA-mediated calcium influx using Fura-2 in chicken embryo neurons. Commercially available NR2B-subunit specific inhibitors Ro 04-5595 and ifenprodil reduced calcium influx with similar IC50 values as in rat neurons whereas spermine potentiated calcium influx, demonstrating functionally active NR2B-containing NMDA receptors, particularly abundant on DIV1 in culture. New potential NR2B-subunit specific inhibitors based on the structure of Ro 04-5595 were synthetized and shown to have a structure-activity relationship to NMDA-mediated calcium influx. Western blot analysis confirmed expression of NR2B in chicken forebrain cultures. Chicken forebrain showed increased NR2B

expression during development from E7 toE18, comparable to rat postnatal development described in literature. We conclude that the chicken embryo nerve cell culture from forebrain is comparable to murine models, and additionally, that it meets the principle of the three R`s in animal experiments (Replacement, Reduction and Refinement).

Disclosures: M.F. Fjelldal: None. **J.E. Jakobsson:** None. **P. Riss:** None. **A. Ring:** None. **I. Sylte:** None. **L.M. Evenseth:** None. **R.E. Paulsen:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.17/D32

Topic: B.02. Ligand-Gated Ion Channels

Title: Development of glutamatergic synapses onto layer 2/3 PV interneurons in the visual cortex during the critical period

Authors: *R. C. FERRER FIERRO[1], A. BAEZ[1], H. HSIEH[3], L. P. WOLLMUTH[2]
[1]Dept. of Neurobio. and Behavior, [2]Dept. of Neurobio. and Behavior & Dept. of Biochem. and Cell Biol., Stony Brook Univ., Stony Brook, NY; [3]Dept. of Surgery, Stony Brook Univ. Hosp., Stony Brook, NY

Abstract: Parvalbumin-expressing (PV) interneurons constitute a subgroup of inhibitory interneurons that regulate circuit dynamics in the brain. The inhibitory role of PV interneurons is mediated by the release of GABA, which is driven in part by the excitatory input that the interneuron receives. Excitatory glutamatergic synapses onto PV interneurons are mediated by both AMPA and NMDA receptors, but compared to pyramidal neurons, they have a small NMDA receptor component and a high proportion of calcium permeable AMPA receptors (GluA2- lacking) to signal excitatory inputs and consequent calcium influx. The normal development of PV interneurons is important for circuit maturation and regulates the critical period for visual plasticity in the visual cortex, but little is known about the dynamics of glutamatergic synapses onto PV interneurons during development. We characterized the properties of excitatory synapses onto PV interneurons in layer 2/3 in the visual cortex during the critical period, specifically addressing how the AMPA receptor (AMPAR) vs. NMDA receptor (NMDAR) components change during development. We found that the overall AMPAR component decreases during the critical period, evidenced by the decrease in the frequency of AMPAR-mediated miniature EPSCs (mEPSCs). On the other hand, the frequency of NMDAR-mediated mEPSCs was highly variable throughout development but showed a notable increase during the critical period. Accordingly, in paired recordings between pyramidal and PV interneurons in layer 2/3, the NMDAR-to-AMPAR ratio is higher during the critical period compared to adult stages. Translaminar glutamatergic connections, from layer 5 to layers 2/3,

showed a dramatically reduced NMDAR-to-AMPAR ratio, suggesting input specific developmental patterns. The developmental changes in NMDAR and AMPAR components during the critical period may influence synaptic integration and calcium dynamics mediated by glutamate receptors and could potentially alter the functional properties of PV interneurons.

Disclosures: R.C. Ferrer Fierro: None. **A. Baez:** None. **H. Hsieh:** None. **L.P. Wollmuth:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.18/D33

Topic: B.02. Ligand-Gated Ion Channels

Support: RS MacDonald Seedcorn award

The Rosetrees Trust Research grant

Title: Modulation of NMDA receptors by group I mGlu receptors delivered via two different pathways

Authors: *S. SYLANTYEV, N. O'NEIL, N. KOMIYAMA, M. STEFAN
Univ. of Edinburgh, Edinburgh, United Kingdom

Abstract: NMDA receptor (NMDAR) hypofunction is one of the key factors provoking schizophrenia development. Another element, whose dysfunction is tightly associated with susceptibility to schizophrenia, is group I metabotropic glutamate receptors (mGluRIs)-mediated signalling. mGluRIs modulate NMDARs via two main pathways. First, metabolic signalling via G-proteins, the inhibition of which is associated with development of schizophrenia. Second, direct potentiation through a Homer1-containing protein interlink. Homer1-knockout animals with destroyed mGluRI-NMDAR interlink exhibit abnormalities consistent with schizophrenia symptoms thus suggesting Homer1 as potential molecule preventing schizophrenia development. Another factor, which destroys Homer-containing molecular interaction, are naturally occurring "short" Homers, such as Homer1a. However, expression of Homer1a protein is upregulated by anti-schizophrenia drugs haloperidole and clozapine. The aim of our study is to resolve this apparent discrepancy and find an approach allowing optimization of anti-schizophrenia drugs action. We set out to research interaction between inputs delivered from mGluRI to NMDR via two different pathways. Based on our pilot data, we hypothesized that mGluRs modulate NMDARs simultaneously via (i) direct molecular interaction through Homer-containing link and (ii) classical G-protein signalling cascade which modify competitively NMDAR functional parameter(s) (opening probability, affinity to agonist, etc.). We investigated this question

through a combination of electrophysiological experimental methods: whole-cell and nucleated patch recordings from control neurones and neurones overexpressing Homer1a. We found, that modulatory impact mediated by Homer proteins is lower than that delivered by G-proteins; fast Homer-delivered effects prevail at short-term time intervals, whereas slow G-protein-delivered input prevails on long-term intervals. Depending on cell type - dentate gyrus granule cells (DGCs) or cerebellar granule cells (CGCs) - Homer-delivered input may potentiate (in DGCs) or antagonize (in CGCs) G-protein delivered input from mGluRs to NMDARs. Our results thus confirmed necessity of the cell-type-specific insight for optimization of anti-schizophrenia drugs action.

Disclosures: S. Sylantyev: None. **N. O'Neil:** None. **N. Komiyama:** None. **M. Stefan:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.19/D34

Topic: B.02. Ligand-Gated Ion Channels

Title: The NMDA receptor modulator NYX-2925 demonstrates therapeutic potential in preclinical models for the treatment of neuropathic pain

Authors: *N. GHOREISHI-HAACK[1], J. PRIEBE[1], J. DUNNING[1], J. BURGDORF[1,2], T. MADSEN[1], M. KHAN[1], C. CEARLEY[1], J. MOSKAL[1,3]
[1]Aptinyx Inc., Evanston, IL; [2]Biomed. Engin., Northwerstern Univ., Evanston, IL; [3]Bimedical Engin., Northwestern Univ., Evanston, IL

Abstract: Aptinyx has developed a novel class of small molecule NMDA receptor modulators with broad applicability across neurologic and psychiatric diseases. Aptinyx's lead compound NYX-2925 is currently in Phase 2 studies for treatment of pain associated with diabetic neuropathy. The present studies examine the effect of NYX-2925 in rat models of neuropathic pain when administered over a wide dose range either as a single dose or two weeks of daily dosing.
The analgesic effect of NYX-2925 was evaluated in the rat chronic constriction injury (CCI) model and the streptozotocin (STZ) model of diabetic neuropathy as measured by mechanical hypersensitivity.
A single oral dose of NYX-2925 produced a rapid-acting (1h post-dosing) and long-lasting (24h and 1 week post-dosing) analgesia in the CCI and STZ models with statistically significant efficacy over vehicle seen between 1-30 mg/kg in the CCI model and 1-10 mg/kg in the STZ model. In contrast, gabapentin (150 mg/kg PO) only produced analgesic effect 1h post-dosing in the both models. Daily oral administrations of NYX-2925 over 14 days results in significant efficacy over vehicle that was sustained throughout the dosing period. The effect of NYX-2925

is both NMDA and AMPA receptor dependent, as systemic co-administration of either the NMDAR antagonist CPP (10 mg/kg, IP) or the AMPAR antagonist NBQX (10 mg/kg, IP) abolishes the analgesic effect of NYX-2925 in the rat CCI model.
These data show that NYX-2925 has therapeutic potential as a daily administered neuropathic pain compound with both rapid acting and long-lasting therapeutic effects that are both NMDAR- and AMPAR-dependent.

Disclosures: **N. Ghoreishi-Haack:** A. Employment/Salary (full or part-time):; Aptinyx Inc. **J. Priebe:** A. Employment/Salary (full or part-time):; Aptinyx Inc. **J. Dunning:** A. Employment/Salary (full or part-time):; Aptinyx Inc. **J. Burgdorf:** A. Employment/Salary (full or part-time):; Aptinyx Inc., NORTHWESTERN UNIVERSITY. **T. Madsen:** A. Employment/Salary (full or part-time):; Aptinyx Inc. **M. Khan:** A. Employment/Salary (full or part-time):; Aptinyx Inc. **C. Cearley:** A. Employment/Salary (full or part-time):; Aptinyx Inc. **J. Moskal:** A. Employment/Salary (full or part-time):; Aptinyx Inc., NORTHWESTERN UNIVERSITY.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.20/D35

Topic: B.02. Ligand-Gated Ion Channels

Support: Takeda Pharmaceutical Company Ltd

Title: Subunit-specific enhancement by 24S-hydroxycholesterol of NMDAR-mediated tonic currents in mouse dentate gyrus granule cells

Authors: *X. WEI[1], T. NISHI[3], S. KONDO[3], H. KIMURA[3], I. MODY[2]
[1]Dept. of Neurol., [2]Dept. of Neurol. and Physiol., The David Geffen Sch. of Med. at UCLA, Los Angeles, CA; [3]Research, Takeda Pharmaceut. Co. Ltd, Fujisawa, Japan

Abstract: 24S-hydroxycholesterol (24HC) is the major metabolic breakdown product of cholesterol in the brain, where it is produced by the enzyme cholesterol 24S-hydroxylase (CYP46A1). 24HC is an endogenous regulator of a variety of neuronal functions. The N-methyl-D-aspartate receptor (NMDAR or GluN) is an important ligand-gated ion channel in the CNS, and its dysfunction is present in several neuropsychiatric disorders, including schizophrenia, depression, autism, Alzheimer's diseases and others. The function of NMDARs is modulated by multiple endogenous and exogenous compounds. One of the endogenous modulators is 24HC, but its mechanism of action onto intact NMDARs in the brain is poorly understood. Here, by using whole-cell patch clamp recordings in dentate gyrus granule cells of mouse brain slices, and pharmacological methods, we recorded isolated NMDAR-mediated (I_{NMDA}) tonic currents and

evoked postsynaptic currents. We found that 1 μM 24HC significantly enhanced tonic I_{NMDA}, but not evoked $I_{NMDA.}$ We next applied PEAQX (a GluN2A antagonist) and Ro25-6891(a GluN2B antagonist) to identify the subtype of GluN modulated by 24HC. Preincubation of the slices for 30 min with PEAQX (300 nM) or Ro25-6891 (1 μM) to block GluN2A and GluN2B respectively, had dramatic effects on the modulatory potential of 24HC. In our preparation where the tonic I_{NMDA} had both GluN2A and GluN2B components, Ro25-6981 blocked the enhancing effect of 24HC on tonic $I_{NMDA,}$ while preincubation with PEAQX had no effect. This indicates that 24HC mostly acts via NMDARs containing GluN2B subunits in intact granule cells. In CYP46A1 knockout mice, where little or no endogenous 24HC is present, 1 or even 10 μM 24HC had no effects on tonic I_{NMDA}, indicating that endogenous 24HC is necessary for maintaining the modulatory function of 24HC on NMDARs in the normal brain. As the subunit composition of NMDARs is different in interneurons (INs) than in principal cells, we also wanted to know whether the subunits present in parvalbumin (PV) INs are also modulated by 24HC. We applied 1 μM 24HC to dentate gyrus PV-INs in mice where PV-INs were labeled with tdTomato (PV cre X Ai14 mice). 24HC had no effect on tonic I_{NMDA} of PV-INs, indicating that the subunits present in these cells (mainly GluN2A and GluN2D) are not targets of 24 HC modulation. Taken together, our results revealed a subunit-specific modulation of NMDARs by 24HC, suggesting a new potential for cholesterol metabolites to influence the function of NMDAR in the brain.

Disclosures: X. Wei: None. **T. Nishi:** A. Employment/Salary (full or part-time):; Takeda Pharmaceutical Company Ltd. **S. Kondo:** A. Employment/Salary (full or part-time):; Takeda Pharmaceutical Company Ltd. **H. Kimura:** A. Employment/Salary (full or part-time):; Takeda Pharmaceutical Company Ltd. **I. Mody:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Supported by a grant from Takeda Pharmaceutical Company Ltd.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.21/D36

Topic: B.02. Ligand-Gated Ion Channels

Support: EKL: CIHR MOP 89825

EKL: Canadian Research Chair

AJR: CIHR

MAB: OGS (Ontario Graduate Scholarship)

Title: How NMDA receptor disruption affects prefrontal cortical neurophysiology essential for executive function

Authors: *M. A. BINKO[1], C. A. MIELNIK[2], A. J. RAMSEY[1,2], E. K. LAMBE[1]
[1]Dept. of Physiol., [2]Dept. of Pharmacol. & Toxicology, Univ. of Toronto, Toronto, ON, Canada

Abstract: The cognitive deficits of schizophrenia include impaired executive function, attention problems and distractibility, indicative of prefrontal cortical disruption. Converging evidence suggests that such cognitive symptoms arise from glutamatergic abnormalities, specifically from N-methyl-D-aspartate (NMDA) receptor hypofunction. Indeed, deficits in executive function are found in transgenic mice with reduced expression of the obligate GluN1 subunit of the NMDA receptor. Here, we examine functional brain changes in the prefrontal cortex of GluN1 knockdown mice relative to wild-type littermate controls. In a tripartite approach, we examined layer 5 pyramidal neurons in acute prefrontal brain slices: (1) measuring basic electrophysiological properties, (2) probing the integrity of synaptic and extra-synaptic NMDA receptor responses, and (3) evoking network activity as an integrated measure of regional brain function. Overall, these experiments paint a striking portrait of the prefrontal cortex in the GluN1 knockdown mice. Many aspects of cellular electrophysiology are largely preserved in the face of lifelong NMDA receptor knockdown, including near-normal NMDA currents to synaptic stimulation. Yet, responses to extra-synaptic NMDA stimulation are clearly disrupted, and electrically evoked network activity is absent under our experimental conditions in the GluN1 prefrontal cortex. As such network activity is thought to be essential for executive function, its disruption in GluN1 knockdown mice would contribute to their observed cognitive deficits. Ongoing work examines the potential enhancement of network activity in GluN1 knockdown and wild-type controls.

Disclosures: M.A. Binko: None. **C.A. Mielnik:** None. **A.J. Ramsey:** None. **E.K. Lambe:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.22/D37

Topic: B.02. Ligand-Gated Ion Channels

Support: NIH NIMH R56 MH107190

NIH 5 F31 MH110096-02

Title: GluN2B subunit selective NMDAR antagonist enhances the excitation/inhibition balance and disinhibits CA1 pyramidal cells only when estrogen is present in female rats

Authors: *A. J. WIDMAN[1], L. L. MCMAHON[2]
[1]Univ. of Alabama at Birmingham (UAB), Birmingham, AL; [2]Dept Cell, Developmental, and Integrative Biol., UAB, Birmingham, AL

Abstract: At low doses, NMDAR antagonists, including ketamine, generate a promising rapid antidepressant response in patients with Major Depressive Disorder (MDD) and in preclinical models with ketamine being the most effective. However, the psychotomimetic properties and abuse potential limit the use of ketamine, making it important to evaluate other NMDAR antagonists. Furthermore, identifying whether mechanistically different NMDAR antagonists impart the same or distinct changes in synaptic network activity in critical brain regions involved in MDD could potentially explain differences in their antidepressant efficacy. Previously, in male rats, we investigated the rapid effects of ketamine and the GluN2B subunit selective NMDAR antagonist, Ro 25-6981, on hippocampal circuits. 1μM ketamine decreases inhibitory input onto pyramidal cells and disinhibits them. In contrast, 1μM Ro 25-6981 had no effect on inhibitory input or pyramidal cell excitability, which correlates with the antidepressant efficacy of each drug. To date, no clinical study has addressed sex differences in response to ketamine, but two preclinical studies show ketamine has a greater antidepressant-like response in ovary-intact females than males (Carrier and Kabbaj, 2013; Franceschelli et al., 2015). Our lab has previously shown that proestrous-like levels of 17β-estradiol (E2) increase GluN2B-containing NMDARs on pyramidal cells (Smith and McMahon, 2006; Vedder et al., 2013). In addition, published studies show acute E2 treatment reduces inhibitory drive onto CA1 pyramidal cells (Huang and Woolley, 2012), suggesting the possibility that a synergistic relationship exists between E2 and NMDAR antagonists in enhancing the excitation/inhibition (E/I) balance in hippocampus. We investigated the rapid effects of ketamine and Ro 25-6981 on hippocampal circuits in ovariectomized female rats following treatment with proestrous-like levels of E2. Results show that 1μM ketamine shifts the E/I balance towards excitation and disinhibits pyramidal cells in female rats independent of E2 levels. However, 1μM Ro 25-6981 only shifts the E/I balance towards excitation and disinhibits pyramidal cells when female rats are treated with proestrous-like E2. These findings indicate the therapeutic dose and antidepressant effectiveness of NMDAR antagonists may fluctuate with varying levels of plasma E2.

Disclosures: A.J. Widman: None. **L.L. McMahon:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.23/D38

Topic: B.02. Ligand-Gated Ion Channels

Support: NIDA R01DA020140 to KSJ

Title: Asymmetrical pH-dependence in the activation of GluN1 and GluN2A N-methyl-D-aspartate receptor subunits

Authors: *N. N. JACKSON, K. S. JONES
Dept. of Pharmacol., Univ. of Michigan Med. Sch., Ann Arbor, MI

Abstract: The N-methyl-D-aspartate receptor (NMDAR) is one of the major subtypes of ionotropic glutamate receptors that mediate excitatory transmission in the central nervous system. NMDAR current is strongly attenuated by extracellular proton concentration (pH), which can vary drastically during pathological states. Structure-function studies have identified several amino acid residues in the transmembrane domains of NMDAR subunits that contribute to pH sensitivity, but the biophysical mechanisms are not fully resolved. The M3 transmembrane domain of the NMDAR forms a gate at the narrowest region of the NMDAR ion channel pore. Here we use the Substituted Cysteine Accessibility Method (SCAM) to examine the influence of extracellular pH on gating-induced translocation of residue A647 in the M3 domain. Cysteine residues were substituted at position A647 and A648 of the GluN1 (GluN1-A7C) and GluN2A (GluN2A-A7C). Mutant NMDAR subunits were expressed in Xenopus oocytes and NMDAR current was measured by two-electrode voltage-clamp electrophysiology. The pH sensitivity of GluN1-A7C- or GluN2A-A7C-containing NMDARs did not differ substantially from WT NMDARs (pH IC_{50} =7.2, WT; 7.3, GluN1-A7C/GluN2A; and 7.1, GluN1/GluN2A-A7C). Bath application of the sulfhydryl-specific compound (2-Aminoethyl) methane-thiosulfonate (MTSEA) did not alter the function of WT NMDARs. We previously showed that bath application of MTSEA alone does not alter the function of NMDARs comprised from A7C-mutant subunits. However, co-application of MTSEA in the presence of agonist dramatically slows channel deactivation (Jones, et al 2002). Here we show MTSEA attenuates pH sensitivity of NMDARs comprised from GluN1-A7C or GluN2A-A7C subunits. MTSEA modifies NMDARs comprised from GluN1/GluN2A-A7C subunits more rapidly and more completely than GluN1-A7C/GluN2A subunits. MTSEA modification increases the net amplitude of current evoked from GluN1/GluN2A-A7C receptors at all pH values tested (pH 6.4-8.4). Curiously, MTSEA modification *increases* the net amplitude of current evoked from GluN1-A7C/GluN2A receptors at acidic values ($pH \leq 7.0$), but *decreases* net amplitude of current evoked at basic values ($pH \geq 7.3$). These differences suggest gating elements in the GluN1 and GluN2A subunits exhibit distinct sensitivities to pH and that protons may induce unique conformations in each subunit. One hypothesis to explain differences in pH sensitivity of the A7C mutants is that cysteine substitution at GluN1-A647C permits the formation of a pH sensitive disulfide bond that is not present in GluN2A-A7C. Data addressing this hypothesis will be presented.

Disclosures: **N.N. Jackson:** None. **K.S. Jones:** None.

Poster

374. NMDA Receptors I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 374.24/D39

Topic: B.02. Ligand-Gated Ion Channels

Support: Boston Children's Hospital Translational Research Program

Title: Static magnetic field modulates cortical synaptic plasticity *In vitro* through an NMDA receptor-dependent pathway

Authors: Y. SUN[1,2,3], P. A. ROSENBERG[1,3], A. PASCUAL-LEONE[4], *A. ROTENBERG[1,2,4]
[1]Dept. of Neurol. and the F.M. Kirby Neurobio. Ctr., [2]The Neuromodulation Program and the Div. of Epilepsy and Clin. Neurophysiol., Boston Children's Hosp., Boston, MA; [3]Program in Neurosci., Harvard Med. Sch., Boston, MA; [4]Berenson-Allen Ctr. for Noninvasive Brain Stimulation, Div. of Cognitive Neurol., Dept. of Neurol., Beth Israel Deaconess Med. Center, Harvard Med. Sch., Boston, MA

Abstract: Background: There is growing evidence that moderate-intensity (1 mT to 1 T) static magnetic fields (SMF) influence cellular membrane channel function in the nervous system. For example, SMF exposure can affect the calcium and sodium current activation kinetics. The mechanism may be explained by the diamagnetic anisotropic properties of membrane phospholipids, which result in a deformation of imbedded ion channels. However, the effects of magnetic fields on neuronal functions are not fully characterized. We examined the SMF influence on neocortical synaptic plasticity in mouse primary motor cortex (M1) in vitro to obtain further insight into SMF effects and underlying mechanisms at the neurotransmitter receptor levels. **Methods:** Field excitatory postsynaptic potentials (fEPSPs) evoked by stimulating the layer 5 to layer 2/3 pathway was recorded in mouse M1 slices using a multichannel electrode array (MED64). Neocortical long-term potentiation (LTP) or depression (LTD), were induced either electrically (200 Hz for LTP; 1 Hz for LTD) or by selective pharmacologic activation of the N-methyl-D-aspartate receptor (NMDAR) or group 1 metabotropic glutamate receptor (mGluR1/5), by NMDA or R,S-dihydroxyphenylglycine (DHPG), respectively. %change of fEPSP slope was monitored before and after LTP/LTD induction with or without SMF (~100mT) exposure which started before LTP/LTD induction and continued for the entire length of the recording. **Results:** Electrically-induced cortical LTP (150.3 ± 12.8% of baseline, $p<0.05$) was eliminated by SMF (105.2 ± 9.3% of baseline, $p>0.05$; and $p<0.05$ as compared to control; n=6). Similarly, electrically-induced cortical LTD (52.9 ± 4.1% of baseline, $p<0.001$) was also significantly attenuated by SMF (78.2 ± 9.1% of baseline, $p>0.05$; and $p<0.05$ as compared to control; n=4). Pharmacologic LTD, generated by brief NMDA (8 μM, 2 min) bath application (52.6 ± 9.8% of baseline, $p<0.01$) was also partially

blocked by SMF (84.4 ± 2.4% of baseline, $p<0.001$; and $p<0.05$ as compared to control; n=6), which suggests an NMDAR-dependent SMF mechanism. In contrast, mGluR-mediated LTD induced by DHPG (50 μM, 5 min) was unaffected by SMF (control: 70.9 ± 7.8% of baseline, $p<0.05$; SMF: 70.8 ± 9.3% of baseline, $p<0.05$ and $p>0.05$ as compared to control; n=3). **Conclusion:** Taken together, our data demonstrate that moderate SMF exposure modulates cortical plasticity in vitro and this modulation is dependent on the NMDAR signaling pathway, and independent of the mGluR1/5 signaling. These findings will be important for understanding the mechanism of therapeutic SMF effects in clinical applications.

Disclosures: Y. Sun: None. **P.A. Rosenberg:** None. **A. Pascual-Leone:** None. **A. Rotenberg:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.01/D40

Topic: B.04. Ion Channels

Support: NIDCD/NIH DC001919

Title: SLK-01, a novel blocker of slack Na+-activated K+ channels

Authors: *D. P. JENKINS[1], V. GRIBKOFF[2], L. K. KACZMAREK[3]
[1]Pharmacol., [2]Intrnl. Medicine: Endocrinol., Yale Univ., New Haven, CT; [3]Yale Univ. Sch. Med., New Haven, CT

Abstract: The sodium-activated K^+ channel (KNa1.1, Slack, Slo2.2) is expressed throughout the brain, and directly couples increases in intracellular sodium to outward hyperpolarizing potassium currents. The Slack channel is thought to mediate a negative feedback system. Surprisingly, gain-of-function mutations in Slack are associated with development of several early onset epileptic encephalopathies. Quinidine, which blocks Slack channels, has recently been used to treat Slack mediated epilepsies. Its low potency and negative side effects profile, however, indicate that development of improved Slack channel blockers is key for the treatment of hyperexcitability-related disorders. SLK-01, originally synthesized for a targeted BK potassium channel activator program, was previously found to have off-target inhibitory activity on Slack channels expressed in Xenopus oocytes. We have now synthesized SLK-01 and have examined its efficacy and preliminary mechanism of action as an inhibitor of Slack currents. Heterologous expression of Slack channels in HEK293T cells showed that SLK-01 was four times more potent than Quinidine. Moreover, analysis of whole-cell current traces and docking simulations indicate that SLK-01 is an open-channel blocker, suggesting that it could be more potent on mutant gain-of-function Slack channels than on wild-type channels. We propose that

development of SLK-01 provides a new tool for investigating ion channel properties, and, critically, establishes a new line of research for discovering novel clinical therapeutics for treating Slack mediated epilepsies.

Disclosures: D.P. Jenkins: None. **V. Gribkoff:** None. **L.K. Kaczmarek:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.02/D41

Topic: B.04. Ion Channels

Title: Lithium may block the delay current (I_D) in olfactory projection neurons

Authors: P. KANTHAKUMAR, *B. I. HYLAND, P. HEYWARD
Univ. of Otago, Dunedin, New Zealand

Abstract: Bipolar disorder is a debilitating neuropsychiatric disease characterized by periodic depressive and manic episodes, associated with altered ion channel gene expression and disturbed brain network activity. Lithium (Li^+) is a first-line treatment for bipolar disorder, but its therapeutic mechanism of action remains unknown. Hypothetical targets for the action of Li^+ range from intracellular signalling messengers to membrane transporters and ion channels. We used whole-cell patch clamp recordings to investigate the effect of Li^+ on the integration of simulated excitatory post synaptic potentials (sEPSPs) by main olfactory bulb output (mitral) cells. The olfactory bulb is a part of the limbic system that is concerned with behaviour and mood and possesses an archetypal cortical modular organization. We found that Li^+ increased mitral cell action potential frequency, decreased after-hyperpolarization (AHP) amplitude, and decreased the decay slope of action potentials. These findings indicate that Li^+ blocks an outward current that plays a role during repolarization and AHP phases of action potentials. Further, Li^+ decreased the delay to action potential generation during a train of sEPSPs, consistent with cumulative inactivation of an outward current. Finally, application of 4-aminopyridine (4-AP) at a concentration that selectively blocks I_D (5 micromolar), reproduced the effects of Li^+ on mitral cell responses to sEPSPs and on action potential phases. Together, these data are consistent with an action of Li^+ on the delay-current (I_D). This current contributes to temporal integration of depolarizing signals due to its exceptionally long kinetics of inactivation and recovery from inactivation, and so can influence cortical network synchrony and spike time precision. Li^+ actions on I_D might therefore influence cortical network activity, contributing to its therapeutic effects in bipolar disorder.

Disclosures: P. Kanthakumar: None. **B.I. Hyland:** None. **P. Heyward:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.03/D42

Topic: B.04. Ion Channels

Support: The Physiological Society

BBSRC

Autifony Therapeutics Ltd

Title: Kv3.3 potassium channel subunits are required to maintain fast synaptic transmission in the auditory brainstem

Authors: A. RICHARDSON[1], S. NEWTON[1], N. PILATI[3], C. H. LARGE[4], V. MARRA[1], *I. D. FORSYTHE[2]
[1]Neuroscience, Psychology & Behaviour, [2]Univ. of Leicester, P.O. Box 138. Leicester, United Kingdom; [3]Autifony Srl Labratories, Verona, Italy; [4]Autifony Therapeut. Limited, London, United Kingdom

Abstract: Voltage-gated potassium channels (Kv) control neuronal excitability, with high voltage-activated Kv3 channels mediating rapid action potential (AP) repolarisation, giving short APs and promoting high frequency firing with temporal precision (*Brew & Forsythe, J. Neurosci. 15, 8011–22, 1995*). Expression across the auditory brainstem nuclei is vital for accurate processing of sound localisation, but specific roles (if any) of the expressed subunits are unknown. Immunohistochemistry shows that of the four Kv3 subunits, Kv3.1 and Kv3.3 dominate in the superior olivary complex (SOC); so functional channels (requiring assembly of 4 subunits) could be as homomers or heteromers. One rationale for multiple subunits could be to influence biophysical properties, another would be to specify trafficking and/or location of the channel to different subcellular compartments.
In vitro patch recording in brainstem slices and *in vivo* auditory brainstem response (ABR) studies were conducted on wildtype and knockout ($Kv3.1^{-/-}$ & $Kv3.3^{-/-}$) CBA/Ca mice to assess synaptic transmission and auditory processing, respectively. ABR recordings using high frequency sound stimuli (24 kHz) were compromised in both knockout mice, while those using low frequency stimuli (12 kHz) were comparable to controls. Voltage clamp recordings during stimulation of the Calyx of Held showed reduced amplitude of synaptic response during high frequency stimulation trains in $Kv3.3^{-/-}$ mice. In contrast, $Kv3.1^{-/-}$ mice showed EPSC failures consistent with axonal action potential transmission failure. Together these data suggest that Kv3.3 is as critical as Kv3.1 for high frequency auditory processing: Kv3.3 has a primary role in

the giant presynaptic terminal, while Kv3.1 predominates in the postsynaptic neuronal soma and in axonal compartments.

Disclosures: **A. Richardson:** A. Employment/Salary (full or part-time):; Auifony Therapeutics Ltd. **S. Newton:** None. **N. Pilati:** A. Employment/Salary (full or part-time):; Autifony Therapeutics Ltd. **C.H. Large:** A. Employment/Salary (full or part-time):; Autifony Therapeutics Ltd. **V. Marra:** None. **I.D. Forsythe:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Autifony Therapeutics Ltd.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.04/D43

Topic: B.04. Ion Channels

Support: NIH grant DC 01919 (L.K.K)

Title: TBK1/ ubiquitination/autophagy pathways are activated by Kv3.3 channels

Authors: ***Y. ZHANG**[1], L. VARELA[2], T. L. HORVATH[2], L. K. KACZMAREK[3]
[1]Yale Univ. Sch. Med., New Haven, CT; [2]Yale Univ., New Haven, CT; [3]Yale Univ., New haven, CT

Abstract: The KCNC3 gene encodes the voltage-dependent potassium channel Kv3.3. These channels are expressed at particularly high levels in Purkinje cells in the cerebellum. Mutations in the KCNC3 gene result in Spinocerebellar ataxia type 13 (SCA 13), a human autosomal dominant disease characterized primarily by degeneration of the cerebellum. Our recent work found that Kv3.3 channel subunits binds Hax-1, an anti-apoptotic protein that is absolutely required for the survival of cerebellar neurons. The interaction of Kv3.3 with Hax-1 leads to the formation of a stable subcortical actin cytoskeleton adjacent to the channels. A human Kv3.3 mutant channel (G592R Kv3.3), which results in late onset SCA13, binds Hax-1 but fails to build the underlying cytoskeleton. Binding of Hax-1 to the channel is required to prevent channel inactivation during sustained depolarization. To probe further how an abnormal interaction between Kv3.3 and Hax-1 leads to cerebellar degeneration, we have now generated G592R knock-in mice, and found that the activity and levels of Tank Binding Kinase-1 (TBK1) are increased four-fold in the cerebellum of G592R Kv3.3 mice over levels in wild type mice. In Kv3.3 expressing cells, Kv3.3 is colocalizes with TBK1, and is immunoprecipitated with this kinase. Depolarization of cells expressing Kv3.3 enhances TBK1 activity. A basal level of TBK1 activity is required to keep Hax-1 bound to Kv3.3, such inhibitors of TBK1 cause the

dissociation of Hax-1 and promote rapid channel inactivation. TBK1 has both serine/ threonine kinase and ubquitin binding domains that allow this enzyme to trigger the binding of ubiquitinated membrane proteins to pre-phagocytic membranes, prior to their internalization into autophagosomes for degradation. By electron microscopy, we find that the enhanced activation of TBK1 in cells expressing the G592R Kv3.3 mutant is associated with increased enhanced formation of intracellular multivesicular bodies and presumed autophagosomes that contain Hax-1. Levels of CD63, a protein marker of multivesicular bodies, are also increased in mutant cells over those in wild type cells. This suggests that Kv3.3 channel activity directly controls the turnover of neuronal components and further suggests that TBK1 activity may regulate neuronal excitability.

Disclosures: Y. Zhang: None. **L. Varela:** None. **T.L. Horvath:** None. **L.K. kaczmarek:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.05/D44

Topic: B.04. Ion Channels

Support: NRSA award F31AA021641

NIAAA R03AA020195

R01AA020992

Tom Calhoon

Title: Behavioral deficits following withdrawal from chronic ethanol are influenced by slo channel function in *C. elegans*

Authors: *J. T. PIERCE, L. SCOTT, S. DAVIS, S. NORDQUIST, G. ORDEMANN
Univ. of Texas at Austin, Austin, TX

Abstract: Symptoms of withdrawal from chronic alcohol use are a driving force for relapse in alcohol dependence. Thus, uncovering molecular targets to lessen their severity is key to breaking the cycle of dependence. Using the nematode *Caenorhabditis elegans*, we tested whether one highly conserved ethanol target, the large-conductance, calcium-activated potassium channel (known as the BK channel or Slo1), modulates ethanol withdrawal. Consistent with a previous report, we found that *C. elegans* displays withdrawal-related behavioral impairments after cessation of chronic ethanol exposure. We found that the degree of impairment is exacerbated in worms lacking the worm BK channel, SLO-1, and is reduced by selective rescue

of this channel in the nervous system. Enhanced SLO-1 function, via gain-of-function mutation or overexpression, also dramatically reduced behavioral impairment during withdrawal. Consistent with these results, we found that chronic ethanol exposure decreased SLO-1 expression in a subset of neurons. In addition, we found that the function of a distinct, conserved Slo family channel, SLO-2, showed an inverse relationship to withdrawal behavior, and this influence depended on SLO-1 function. Together, our findings show that modulation of either Slo family ion channel bidirectionally regulates withdrawal behaviors in worm, supporting further exploration of the Slo family as targets for normalizing behaviors during alcohol withdrawal.

Disclosures: J.T. Pierce: None. **L. Scott:** None. **S. Davis:** None. **S. Nordquist:** None. **G. Ordemann:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.06/D45

Topic: B.04. Ion Channels

Support: NIH Grant 1R01NS092705

Title: The potassium channel Kv4.2 regulates seizure susceptibility in mouse models

Authors: *D. TIWARI[1], L. SCHROEDER[1], R. DANZER[1], A. BUNK[1], S. C. DANZER[2,3], C. GROSS[5,4]
[2]Dept Anesthesia, [1]Cincinnati Children's Hosp. Med. Ctr., Cincinnati, OH; [3]Dept. of Anesthesiol., [4]Dept. of Pediatrics,, Univ. of Cincinnati, Cincinnati, OH; [5]Neurol., Cincinnati Childrens Hosp. Med. Ctr., Cincinnati, OH

Abstract: The voltage-gated potassium channel Kv4.2 is a critical regulator of dendritic excitability in the hippocampus, and is crucial for dendritic signal integration. Kv4.2 mRNA and protein expression as well as function are reduced in several genetic and pharmacologically induced rodent models of epilepsy. However, so far it is not known if reduced Kv4.2 is just an epiphenomenon of epilepsy, or if it causes neuronal hyperexcitability and increased seizure susceptibility. Our previous studies suggest that microRNA-induced downregulation of Kv4.2 contributes to seizure development in a mouse model of kainic-acid induced *status epilepticus*. In the present study, we used Kv4.2 heterozygous mice and adult-onset manipulation of hippocampal Kv4.2 expression in mice to assess Kv4.2's role in regulating hyperexcitability and neuronal morphology in the brain. Western blot and quantitative real-time PCR analyses confirmed a significant reduction of Kv4.2 protein and mRNA levels in the Kv4.2 heterozygous mice. Using EEG analyses, we show that the latency to onset of kainic acid-induced seizures is

significantly shortened in Kv4.2 heterozygous mice compared to wildtype littermates. In addition, a significant increase in total EEG power was observed. No changes in anxiety or overall activity could be detected. Preliminary studies suggest that intrahippocampal CA1 injection of lentivirus overexpressing Kv4.2 delays seizure onset compared to GFP-expressing controls. Overall, these results show that Kv4.2 expression levels regulate seizure onset and severity. In the future, manipulation of Kv4.2 expression and function may be used to alter seizure susceptibility in epilepsy.

Disclosures: D. Tiwari: None. **L. Schroeder:** None. **R. Danzer:** None. **A. Bunk:** None. **S.C. Danzer:** None. **C. Gross:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.07/D46

Topic: B.04. Ion Channels

Support: MOST 105-2320-B-010-016-MY3 (Taiwan)

Title: Regulation of ether-à-go-go potassium channel expression by RING E3 ubiquitin ligases

Authors: *Y.-C. FANG[1,2], C.-Y. TANG[2], C.-J. JENG[1]
[1]Inst. of Anat. & Cell Biol., Natl. Yang-Ming Univ., Taipei, Taiwan; [2]Dept. of Physiol., Natl. Taiwan Univ., Taipei, Taiwan

Abstract: The *ether-à-go-go* (Eag) potassium channel belongs to the superfamily of voltage-gated potassium channel. In mammals, the expression of Eag potassium channels is neuron-specific and is widely distributed over various brain regions. Two Eag isoforms have been identified in rat, Eag1 and Eag2. We have applied the yeast two-hybrid screening system to identify rat Eag1 (rEag1)-interacting proteins from a rat brain cDNA library. Here we report the identification and characterization of RING E3 ubiquitin ligases as rEag1-binding partners. The physical interaction between RING E3 ubiquitin ligases and rEag1 were confirmed with GST pull-down assays and co-immunoprecipitation, which was in agreement with their co-localization with rEag1 at synaptic regions in cultured neurons. rEag1 protein expression level was significantly increased when endogenous RING E3 ubiquitin ligase expression in HEK293T cells was knocked down. Over-expression of RING E3 ubiquitin ligases led to reduced protein level, enhanced ubiquitination, accelerated protein turn-over, and decreased current density of rEag1 channels. The RING E3 ubiquitin ligase-induced rEag1 protein reduction was rescued by the proteasome inhibitor MG132 and the lysosomal inhibitor chloroquine. Together, our results indicate that RING E3 ubiquitin ligases contribute to both endoplasmic reticulum and peripheral protein quality control of rEag1 channels.

Disclosures: **Y. Fang:** None. **C. Tang:** None. **C. Jeng:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.08/D47

Topic: B.04. Ion Channels

Support: NIH Grant FI2GM120004

Title: A Cav2.3-Kv4.2 ion channel complex imparts local Ca^{2+} enhancement of A-type K^+ current

Authors: ***J. G. MURPHY**, L. LIN, E. E. GRAY, J. GUTZMANN, J. HU, D. A. HOFFMAN
Natl. Inst. for Child Hlth. and Human Develop., NIH, Bethesda, MD

Abstract: The rapidly inactivating A-type K^+ current mediated by Kv4.2 regulates action potential repolarization, subthreshold dendritic excitability, and synaptic plasticity in CA1 pyramidal neurons of the hippocampus. Kv4.2 channels assemble with the auxiliary subunits Dipeptidyl aminopeptidase-like proteins (DPP6/10) and members of the K^+ channel interacting protein family (KChip). KChips bind to the intracellular N-terminus of Kv4.2 and function to increase channel expression and function. Importantly, KChips bear 4 EF-hand motifs that are hypothesized to bestow Ca^{2+} regulation upon Kv4.2 channels. Recent evidence suggests that Kv4.2 function is regulated by R-type Ca^{2+} channel (Cav2.3) mediated Ca^{2+} entry in a KChip-dependent manner. However, the molecular nature of a Kv4.2-Cav2.3 interaction has not been explored.
Here we present evidence for a direct Kv4.2-Cav2.3 interaction that imparts Ca^{2+} regulation upon Kv4.2. We first identified Cav2.3 and Kv4.2 as interaction partners in a tandem affinity purification mass spectrometry screen using Kv4.2 as bait. We confirmed the interaction by coimmunoprecipitation of a Kv4.2-Cav2.3 complex from both HEK293 cells and hippocampal lysates and by immunogold labeling and electron microscopy. Next, photobleaching and sensitized emission Förster resonance energy transfer (FRET) experiments provided the evidence for a direct interaction between Cav2.3 and Kv4.2 that is independent of auxiliary subunits. Kv4.2-CFP fluorescence recovery after photobleaching (FRAP) demonstrated a small mobile fraction of Kv4.2 as we previously published. By comparison, Cav2.3-YFP has a large mobile fraction. Coexpression of Cav2.3-YFP and Kv4.2-CFP dramatically decreases the mobility of Cav2.3-YFP, suggesting Kv4.2 mediates recruitment of Cav2.3-YFP into a relatively immobile protein complex. To determine whether a Cav2.3-containing complex regulates Kv4.2 function, we measured K^+ currents under whole cell voltage clamp in HEK293FT cells expressing Kv4.2 and KChip2a alone or with Cav2.3. Under these conditions, K^+ currents are increased ~25%, while replacing EGTA with BAPTA in the patch pipette or extracellular application of the

Cav2.3 antagonist Ni^{2+} reversed this increase. Additionally, whole cell recordings in CA1 hippocampal neurons revealed a decrease in A-type K^+ current density in Cav2.3 knockouts when compared to wild-type mice. Taken together, these results support a direct Cav2.3-Kv4.2 interaction in which Ca^{2+} entry through Cav2.3 increases Kv4.2 function in hippocampal neurons. Ongoing efforts are exploring how the Cav2.3-Kv4.2 interaction regulates excitability in the hippocampus.

Disclosures: J.G. Murphy: None. **L. Lin:** None. **E.E. Gray:** None. **J. Gutzmann:** None. **J. Hu:** None. **D.A. Hoffman:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.09/D48

Topic: B.04. Ion Channels

Support: NIH intramural research program

Title: Activity dependent phosphorylation of Kv4.2

Authors: *J.-H. HU, G. TABOR, Y. LIU, D. A. HOFFMAN
NICHD, Bethesda, MD

Abstract: Kv4.2, the prominent A-type voltage-gated potassium channel expressed in hippocampal CA1 pyramidal neuron dendrites, plays an important role in regulating the back-propagating action potentials and limiting the propagation of local dendritic spikes (Hoffman DA, 1997; Lai HC and Jan LY. Nat Rev Neurosci 2006). Kv4.2 control of dendritic excitability impacts neuronal plasticity and contributes to learning and memory (Lugo JN, 2015). Kv4.2 is regulated by its auxiliary subunits (DPP6/10 and KChIPs) and phosphorylation (Jerng HH, 2004). Previous findings showed that Kv4.2 can be phosphorylated at T602, T607 and S616 by ERK in vitro (Adams JP, 2000). However, it is still unknown if phosphorylation is regulated in brain and what function it has. Here, we report that Kv4.2 is dynamically phosphorylated at T607 in mouse brain. We first identified endogenous Kv4.2 phosphorylation sites in cultured hippocampal neurons using tandem affinity purification followed by mass spectrometry analysis. We found that Kv4.2 is phosphorylated at ERK sites and that T607 phosphorylation is increased when neurons were stimulated with AMPA. To further study Kv4.2 phosphorylation, we characterized specific antibodies against phosphor-T602 and phosphor-T607 using Kv4.2 mutations. We then detected phosphor-T602 and phosphor-T607 of Kv4.2 in mouse brain using the characterized antibodies. Interestingly, T607 phosphorylation was induced by seizure triggered by pentylenetetrazol administration, while T602 phosphorylation remained unchanged. We also found that about 10% of Kv4.2 is phosphorylated at T602 and 5% at T607 in mouse

brain, and that dual phosphorylation at T602 and T607 exists in mouse brain. To further investigate the role of T607 phosphorylation, we generated a knockin mouse with T mutated to A at 607 site using Crispr-Cas9 techniques. These mice will be analyzed for learning and memory behaviors. Taken together, these data support the idea that Kv4.2 is dynamically phosphorylated at T607 in brain and suggest it may play an important role in regulation of brain function.

Disclosures: J. Hu: None. **G. Tabor:** None. **Y. Liu:** None. **D.A. Hoffman:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.10/D49

Topic: B.04. Ion Channels

Support: Intramural Research Program of the Eunice Kennedy Shriver National Institute of Child Health and Human Development

Title: DPP6 loss results in behavioral impairments in recognition, learning and memory

Authors: L. LIN[1], J. G. MURPHY[2], R.-M. KARLSSON[3], R. S. PETRALIA[4], H. A. CAMERON[3], *D. A. HOFFMAN[1]
[2]NIGMS, [3]NIMH, [1]NIH, Bethesda, MD; [4]Advanced Imaging Core, NIDCD/NIH, Bethesda, MD

Abstract: DPP6 plays an important role as an auxiliary subunit of Kv4-mediated A-type K+ channels. However, in our recent previous studies, we found a novel role for DPP6 in synaptic development and function, which may be independent of its role as a K+ channel auxiliary subunit (Lin et al, 2013, 2014). In this study, we investigated the behavioral consequences of DPP6 loss.
We found that DPP6-KO mice were impaired in hippocampal dependent novel object recognition and object's spatial location tasks. Results from the Morris water maze, showed that DPP6-KO mice exhibit slower learning and reduced memory performance. In the T-Maze task, the success rate of WT mice increased over the course of the training whereas DPP6-KO mice showed no significant improvement throughout the training. The locomotor activity of DPP6-KO mice are not significantly difference than WT mice in both light and dark phases in the home cage task. Taken together these results show that impaired synaptic development, results in spatial learning and memory deficiencies in adult DPP6-KO mice. Ongoing experiments are aimed at determining the molecular mechanisms behind the synaptic developmental changes.

Disclosures: L. Lin: None. **J.G. Murphy:** None. **R. Karlsson:** None. **R.S. Petralia:** None. **H.A. Cameron:** None. **D.A. Hoffman:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.11/D50

Topic: B.04. Ion Channels

Title: SUMOylation of the mouse voltage-gated potassium channel Kv4.2 decreases the maximal conductance of the A-type potassium current (I_A)

Authors: *M. A. WELCH[1], L. A. FORSTER[2], D. J. BARO[1]
[1]Biol., [2]Neurosci. Inst., Georgia State Univ., Atlanta, GA

Abstract: Small ubiquitin-like modifier (SUMO) is ~100 amino acid peptide post-translationally added to a lysine residue within a SUMOylation consensus site on a target protein. SUMOylation usually facilitates protein-protein interactions and is an important mediator of neuronal function. SUMOylation is necessary for several forms of synaptic plasticity and intrinsic plasticity mechanisms could also rely on SUMOylation because it has been shown that ion channel surface expression can vary with the extent of channel SUMOylation. The voltage-gated potassium channel Kv4.2 mediates I_A. In hippocampal neurons, Kv4.2 surface expression can be regulated in an activity dependent manner such that there is a protein kinase A (PKA) dependent internalization of Kv4.2 channels following synaptic stimulation or during long-term potentiation (LTP). Our data suggest that SUMOylation could be involved in Kv4.2 channel internalization. Using the SUMOplot analysis program, we identified several putative SUMOylation sites on the Kv4.2 channel including one high probability site (79%) that is also present in the Kv4.3 channel. To test the hypothesis that SUMOylation at this site regulates the surface expression of Kv4.2 channels, a GFP-tagged mouse Kv4.2 channel was stably expressed in human embryonic kidney (Hek) cells (Hek-Kv4.2g). Baseline SUMOylation was or was not increased by transiently transfecting Hek-Kv4.2g cells with SUMO and the SUMO conjugating enzyme Ubc9. Whole cell patch clamp recordings showed that when baseline SUMOylation was enhanced, I_A maximal conductance was significantly reduced (37.6±3.7μS vs. 24.1±2.1μS, $p<0.05$, t-test). The voltage dependence of activation and inactivation and the kinetics of inactivation were unaltered. Changing lysine to arginine within the high probability SUMO consensus site prevented the decrease (27.0±2.7μS vs. 27.9±2.8μS, $p=0.8$, t-test). These data suggest that the Kv4.2 channel is SUMOylated at the high probability consensus site in Hek-Kv4.2g under baseline conditions and that increasing SUMOylation decreases Kv4.2 surface expression. Immunoprecipitation experiments with an anti-GFP antibody followed by western blotting with antibodies against SUMO and Kv4.2 indicated that Kv4.2 channels were SUMOylated in Hek-Kv4.2g cells. Currently we are quantifying SUMOylation and surface expression of wild type and mutant Kv4.2 channels in a heterologous expression system. Similar experiments are being performed with the mouse Kv4.3 channel and we are examining Kv4.2 and Kv4.3 channel SUMOylation in

the mouse CNS. This work will enhance our understanding of SUMO's role in regulating Kv4.2 and Kv4.3 surface expression.

Disclosures: M.A. Welch: None. **L.A. Forster:** None. **D.J. Baro:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.12/D51

Topic: B.04. Ion Channels

Support: NIH grant NS078152

NIH grant NS096296

Title: BK channels are structurally and functionally coupled with NMDA receptors and postsynaptically regulate synaptic transmission and plasticity

Authors: X. GUAN[1], J. ZHANG[1], Q. LI[1], A. MEREDITH[2], H.-L. PAN[1], *J. YAN[1]
[1]Dept. of Anesthesiol. and Perioperative Med., Univ. of Texas MD Anderson Cancer Ctr., Houston, TX; [2]Dept. of Physiol., Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: The large conductance calcium- and voltage-activated potassium (BK) channel is widely expressed in the central nervous system. The physiological roles of BK channels and the calcium sources of their activation in mammalian brains remain not well-understood. Our proteomic and biochemical analyses of BK channels and the calcium-permeable NMDA receptors have found that they form protein complexes in whole brain and various brain regions, consisting of the obligatory BK channel alpha (BKα) and GluN1 subunits and at least the regulatory GluN2A and GluN2B subunits. We examined the cellular colocalization and functional coupling of BK channels to NMDA receptors in mice hippocampal dentate gyrus. We found that they colocalized within nanodomains in dentate gyrus and BK channels were robustly activated by NMDAR-mediated Ca^{2+}-influx in granule cell somas. We observed that blockade of BK channels by paxilline increased the amplitude of excitatory postsynaptic potentials (EPSPs) of granule cells evoked by stimulation in the perforant path-granule cell synapses, which was prevented by blockade of postsynaptic NMDARs or BK channels via pipette delivery of MK-801 or paxilline or by genetic ablation of BKα. We also found that blockade of postsynaptic BK channels and genetic ablation of BKα both abolished long-term potentiation (LTP) of granule cells. We thus inferred that by complex formation and functional coupling with NMDA receptors, postsynaptic BK channels are critically involved in synaptic transmission and LTP in hippocampal dentate gyrus.

Disclosures: **X. Guan:** None. **J. Zhang:** None. **Q. Li:** None. **A. Meredith:** None. **H. Pan:** None. **J. Yan:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.13/D52

Topic: B.04. Ion Channels

Support: NIH Grant DE018661

NIH Grant DE023090

Title: Temperature-sensitive outward currents through two-pore domain K^+ channels in nociceptive-like primary afferent neurons

Authors: *V. VIATCHENKO-KARPINSKI, F. EROL, J. LING, J. G. GU
Anesthesiol., Univ. of Alabama At Birmingham, Birmingham, AL

Abstract: Two-pore domain K^+ channels (K2P) play important roles in setting resting membrane potentials and neuronal excitability. Activity of K2P channels has been found to be highly temperature-dependent, raising a possibility that these channels may be involved in setting neuronal excitability at different temperatures in primary afferent neurons. In the present study we seek to characterize K2P channels in nociceptive type of primary afferent neurons in rat dorsal root ganglia (DRG) to determine their temperature sensitivity and pharmacological properties. Under the condition when cations flowing through voltage-gated ion channels were inhibited, we measured outward currents to determine K2P channel activity in nociceptive-like DRG neurons acutely dissociated from adult rats. The outward currents were recorded using whole-cell patch-clamp recordings with a Cs^+-based intracellular recording solution, which had reversal potentials near -80 mV. Based on the current amplitude and thermal sensitivity of the outward currents, cells could be classified into three types. In the first type (type I), outward currents had large amplitude at the temperature of 22^{o}C, and the amplitude of the outward currents increased when temperature increased to 30^{o}C and reduced when temperature dropped to 14^{o}C. In the second type (type II), outward currents had large amplitude at the temperature of 22^{o}C, and the amplitude of the outward currents did not increase when temperature increased to 30^{o}C but reduced when temperature dropped to 14^{o}C. In the third group (type III), outward currents had small amplitude at the temperature of 22^{o}C, and the amplitude of the outward currents did not increase when temperature increased to 30^{o}C but increased when temperature dropped to 14^{o}C. Pharmacological properties of the temperature sensitive outward currents in these three types of cells were examined. Outward currents in type 1 cells but not type 2 and type 3 cells could be inhibited by PGF2α, a compound that has recently been shown to have effects on

K2P channels. Gd^{3+} and riluzole, two compounds that can inhibit K2P channels, were also tested and they appeared to produce inhibitory effects on the outward currents of type I and type II cells. Taken together, this study may suggest that different types of K2P channels are present in functionally distinct groups of nociceptive neurons, and their functions in setting nociceptive neuron excitability need to be determined in future studies.

Disclosures: V. Viatchenko-Karpinski: None. **F. Erol:** None. **J. Ling:** None. **J.G. Gu:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.14/D53

Topic: B.04. Ion Channels

Title: 17β-Estradiol potentiates TREK-1 activity through G protein-coupled estrogen receptor

Authors: *N. CHOUDHURY, S. K. SIKDAR
Mol. Biophysics Unit, Indian Inst. of Sci., Bangalore, India

Abstract: TREK-1 ($K_{2P}2.1$), a two-pore domain leak potassium channel, is implicated in controlling the excitability in neurons owing to its current properties. It is involved in physiological and pathophysiological processes such as pain, anesthesia, epilepsy, ischemia, and depression. Activity of TREK-1 is modulated by various chemical and physical stimuli including membrane receptor - coupled second messengers. Previous studies have shown an inhibitory effect of G_s and G_q Protein - Coupled Receptors on TREK-1 channel activity. However, much less is known about GPCR mediated activation of TREK-1. In the present study we investigate the modulation of TREK-1 by 17β-Estradiol, a reproductive hormone as well as a neuromodulator, using cell-attached patch clamp recordings of TREK-1 ion channel currents. 17β-Estradiol can activate several signaling pathways in the cell on binding to its membrane bound receptor G - Protein coupled Estrogen Receptor (GPER). Experiments in HEK293 cells expressing GPER and TREK-1 indicate about 4 - fold increase in TREK-1 channel activity within minutes of 17β-Estradiol application in a pertussis toxin - sensitive manner. The involvement of GPER is confirmed with its agonist G-1 and antagonist G-15. Pharmacological inhibition of signaling pathways indicate involvement of the α - subunit of G_i with the resulting decrease in cAMP leading to reduced activity of Protein Kinase A and thus activation of TREK-1. Mutational studies indicate the serine residues in the C-terminal domain of TREK-1 to be the sites of action of 17β-Estradiol through GPER. The study reveals the potentiating role of 17β-Estradiol on TREK-1 channel activity through GPER by decreasing phosphorylation in the C-terminal domain of TREK-1. This may be one of the mechanisms by which 17β-Estradiol controls cellular excitability and acts as a neuroprotectant.

Disclosures: **S.K. Sikdar:** A. Employment/Salary (full or part-time):; Ministry of Human Resource Development, Govt. of India. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Department of Biotechnology, Govt. of India, Ministry of Human Resource Development, Govt. of India.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.15/D54

Topic: B.04. Ion Channels

Support: NIDCD/NIH-2R01DC001919-24

FRAXA Foundation-R13005

Title: The functional modulation of sodium-activated potassium (Slack) channels by Phactr1

Authors: *S. R. ALI, L. KACZMAREK
Dept. of Pharmacol., Yale Univ., New Haven, CT

Abstract: The Slack gene encodes sodium-activated potassium channels that are abundantly expressed in the central nervous system. Human mutations alter the function of Slack channels, resulting in epilepsy and intellectual disability. Most of the disease-causing mutations are located in the extended cytoplasmic C-terminus of Slack channels and most result in increased Slack current. Previous experiment using a yeast two-hybrid system indicate that the C-terminus of Slack channels binds a number of cytoplasmic signaling proteins. One of these is Phactr1, a protein that is believed to target protein phosphatase 1 (PP1) to its phosphoprotein substrates. Phactr1 is also known to be an actin-binding protein. We have now found by co-immunoprecipitation that all three components, Phactr1, PP1 and actin exist in a complex with Slack channels. Upon activation of protein kinase C, Phactr1 dissociates from the Slack channel. Conversely, pharmacological activation of PP1 increased the binding of Phactr1 to Slack channels. We have also monitored the association/dissociation of Slack channels with Phactr1 using a fluorescence resonance energy transfer (FRET) assay. As in the biochemical experiments, activation of PP1 stimulated the association of Phactr-1 with the channel. In parallel experiments using whole-cell patch clamp recordings of Slack currents we found that co-expression of Phactr1 produced a suppression of Slack current. Because phosphorylation of Slack channels at serine residue 407 produces an increase in current, our findings are consistent with the hypothesis that Phactr1 recruitment of PP1 to the channel results in dephosphorylation and loss of current amplitude. Our findings suggest that targeting Slack-Phactr1 interactions may be helpful in developing novel therapies for brain disorders associated with the malfunction of Slack channels.

Disclosures: **S.R. Ali:** None. **L. Kaczmarek:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.16/D55

Topic: B.04. Ion Channels

Support: NIH Grant 5R01HD067517

NIH Grant 5R01DC001919

Swebilius Foundation

Title: Mouse model of slack-associated epilepsy

Authors: *I. H. QURAISHI[1], M. MERCIER[2], Y. ZHANG[3], D. P. JENKINS[4], K. P. MANGAN[5], L. K. KACZMAREK[2]
[1]Dept. of Neurol., [2]Yale Sch. of Med., New Haven, CT; [3]Yale Univ. Sch. Med., New Haven, CT; [4]Pharmacol., Yale Univ., New Haven, CT; [5]Cell. Dynamics, Intl., Madison, WI

Abstract: Mutations in the potassium channel Slack (also called KCNT1 or KNa1.1) lead to various types of epilepsy including autosomal dominant frontal lobe epilepsy (ADNFLE) and epilepsy of infancy with migrating focal seizures (EIMFS). In the past few years, it has become clear that this is an important epilepsy gene, with over 20 epilepsy-causing mutations identified. Slack is important in the synchrony of high frequency neuronal activity, survival from electroshock seizures, and in the regulation of neuronal protein translation. Almost all the mutations that have been tested to date by heterologous expression in Xenopus oocytes cause an increase in the sodium-activated potassium current. Using CRISPR-Cas9 we have now developed a mouse model of Slack-associated epilepsy with the R455H mutation, which was identified in patients with epilepsy of infancy with migrating focal seizures (EIMFS). Homozygotes have low survival. Heterozogotes have spontaneous subclinical and tonic-clonic seizures on video EEG monitoring as well as interictal epileptiform discharges. Latencies for pentylenetetrazole-induced seizures are substantially reduced in heterozygous mice. The mice have deficits in procedural learning, as was also seen in Slack knockout mice. In addition to the mouse model, we evaluated neurons derived from induced pluripotent stem cells and engineered to have Slack P924L, another mutation associated with EIMFS. Whole cell voltage clamp recordings confirmed a gain of function of the sodium-activated current. In addition, multielectrode array recordings demonstrated an increase in bursting behavior. We have begun screening for selective modulators of the Slack channel as there are currently no specific drugs available. We identified a potent activator as well as the first state-dependent blocker of the

channel. Taken together, these results will form the framework for identifying the mechanisms of Slack-associated epilepsy and for targeted anti-epileptic drug discovery.

Disclosures: **I.H. Quraishi:** None. **M. Mercier:** None. **Y. Zhang:** None. **D.P. Jenkins:** None. **K.P. Mangan:** A. Employment/Salary (full or part-time):; Fujifilm/Cellular Dynamics International. **L.K. Kaczmarek:** F. Consulting Fees (e.g., advisory boards); Praxis Pharmacueticals.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.17/D56

Topic: B.04. Ion Channels

Support: P30ES005022

R21ES027119

Title: The effects of organophosphate flame retardants on the M-current in NPY neurons in adult mice

Authors: *G. VAIL[1], T. A. ROEPKE[2]
[1]Toxicology, Rutgers Univ., New Brunswick, NJ; [2]Animal Sci., Rutgers, The State Univ. of New Jersey, New Brunswick, NJ

Abstract: The hypothalamus regulates many important biological functions such as reproduction, thermoregulation, and energy balance. Orexigenic NPY neurons located in the arcuate nucleus (ARC) of the hypothalamus play an essential role in energy homeostasis. Disruption of this balance can cause metabolic disease states such as obesity and anorexia. EDCs such as organophosphate flame retardants (OPFR) are a potential cause of hypothalamic disruption as they accumulate in human tissues and impinge on endogenous nuclear receptors such as ERα and PPARγ. Previously, we demonstrated that OPFR oral treatment in adult mice decreased ARC expression of neuropeptide Y and increased growth hormone secretagogue receptor (GHSR) and KCNQ channel subunit expression in a sex-dependent manner. We hypothesized that these effects would augment the activity of the M-current in NPY neurons leading to a suppression of NPY excitability and a reduction in food intake. We will use voltage clamp, whole-cell patch-clamp to record the M-current in NPY neurons utilizing selective KCNQ channel blocker XE991 with standard activation and deactivation protocols from male and females after OPFR treatment. These experiments will be coupled with single cell-type quantitative real-time PCR to measure expression of neuropeptides, GHSR, and KCNQ subunits in NPY pools. Our data suggests that adult exposures to EDC especially flame-retardants may

impact the hypothalamic melanocortin neurocircuitry leading to dysregulation of energy homeostasis.

Disclosures: G. Vail: None. **T.A. Roepke:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.18/D57

Topic: B.04. Ion Channels

Support: HIH DC01919

NIDCD RO1DC00273

Title: Loss of Kv1.3 potassium channels impairs auditory function

Authors: *L. EL-HASSAR[1], L. SONG[2], V. R. GAZULA[1], D. NAVARATNAM[3], J. SANTOS-SACCHI[2], L. K. KACZMAREK[1]
[1]Pharmacol. department SHMB 309, Yale Univ. Sch. of Med., New Haven, CT; [2]Surgery, [3]Neurol., Yale Sch. of Med., New Haven, CT

Abstract: Kv1.3 is a voltage-dependent potassium channel involved in various physiological functions such as cell volume regulation, proliferation, and insulin signaling. Within the central nervous system, deletion of the Kv1.3 gene from mitral cells of the olfactory bulb has been found to increase dramatically the sensitivity of the olfactory system. To test the effects of loss of Kv1.3 on another sensory system, we have analyzed its expression in the auditory system. We used an anti-Kv1.3 antibody to localize these channels to bouton-like structures on inner and outer hair cells within the cochlea. Specificity of staining was confirmed by the finding that immunostaining of these structures is completely absent in Kv1.3 $^{-/-}$ mice. Our group has also shown that Kv1.3 channels are present in presynaptic terminals of the medial nucleus trapezoid body (MNTB) within the auditory brainstem. We next combined *in vivo* with *in vitro* approaches to test the contribution of these channels to auditory function in the peripheral and central auditory system. Measurements of auditory brainstem responses (ABR) show that auditory thresholds are elevated in Kv1.3 $^{-/-}$ mice over those in wild type (WT) mice. Latencies of peaks I, II and IV, which represent transfer of auditory information from the cochlea to midbrain, are prolonged in Kv1.3 $^{-/-}$ mice. In addition, we found a desynchronization of ABR waves in Kv1.3 $^{-/-}$ mice suggesting an alteration of synaptic transmission and changes in spike fidelity within these auditory pathways. To further investigate the mechanisms of these alterations in ABR waves, we carried out patch-clamp recordings of the high-fidelity calyx of Held/MNTB synapse. Our results show that lack of Kv1.3 channels significantly increases the firing frequency of

presynaptic terminals (Calyxes of Held) in response to square pulses of injected currents. In contrast, no difference between wild type and Kv1.3$^{-/-}$ animals was detected in postsynaptic MNTB neurons. This result suggests that loss of Kv1.3 channels impairs auditory function primarily by influencing the properties of presynaptic terminals. Ongoing experiments are characterizing synaptic transmission between the calyx of held and MNTB principal neurons in both Kv1.3 $^{-/-}$ and wild type mice.

Disclosures: L. El-Hassar: None. **L. Song:** None. **V.R. Gazula:** None. **D. navaratnam:** None. **J. Santos-Sacchi:** None. **L.K. Kaczmarek:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.19/D58

Topic: B.04. Ion Channels

Support: NIH grant AG038910

NIH grant NS098328

NIRG-10-174150

Title: Upregulation of microglial Kv1.3 potassium channels in neuroinflammatory disorders

Authors: *H. M. NGUYEN[1], Y.-J. CHEN[2], H. WULFF[2], I. MAEZAWA[3], L.-W. JIN[4]
[1]Pharmacol., Univ. of California Davis, Davis, CA; [2]Pharmacol., UC Davis, Davis, CA; [3]M.I.N.D. Inst, UC Davis, Sacramento, CA; [4]M.I.N.D. Inst. UC Davis, Davis, CA

Abstract: Microglia effector functions are widely associated with specific phenotypes, which are referred to as M1 and M2, in response to cues from the surrounding brain microenvironment. While classically activated M1 microglia release pro-inflammatory cytokines and neurotoxic molecules and have been associated with neurological damage in ischemic stroke, Alzheimer's and Parkinson's diseases, alternatively activated M2 microglia exhibit beneficial immunological effector functions such as phagocytosis of debris and release of anti-inflammatory and neurotrophic factors. We have previously identified three major types of K^+ channels in M(LPS)- and M(IL-4)-differentiated cultured primary mouse microglia based on their biophysical and pharmacological fingerprints: Kv1.3, Kir2.1, and KCa3.1. However, whether these K^+ channels are preferentially expressed in M1-like microglia in diseases *in vivo* is unknown. We first demonstrated higher Kv1.3 current densities by whole-cell patch-clamp electrophysiology in acutely isolated CD11b-positive microglia from brains of mice injected with the pro-inflammatory stimulus lipopolysaccharide **(**LPS). The AD-related toxin amyloid-β oligomer (AβO) also induced increased Kv1.3 in mice when injected but with the addition of the Kir2.1

channel expression, suggesting this stimulus involves a different activation pathway. These microglia also exhibit increased expression of the pro-inflammatory mediators TNF-α, IL-1β, IL-6 and iNOS, confirming that upregulation of Kv1.3 channels are indeed occurs in activated microglia. To demonstrate its physiological relevance, we showed elevated Kv1.3 current expression from microglia from the infarcted hemisphere of mice subjected to middle cerebral artery occlusion (MCAO) and brains of 5xFAD mice, animal models of ischemic stroke and AD, respectively. Importantly, using immunohistochemistry, we observed strong Kv1.3 expression on microglia/macrophage in human stroke and AD biopsies. These findings validate Kv1.3 as potential target for inhibitors as potential therapeutic agents for preferentially inhibiting pro-inflammatory M1 microglia functions in ischemic stroke and other neurological diseases such as Alzheimer's and Parkinson's disease.

Disclosures: H.M. Nguyen: None. **Y. Chen:** None. **H. Wulff:** None. **I. Maezawa:** None. **L. Jin:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.20/D59

Topic: B.04. Ion Channels

Support: RFBR 16-04-00490

Mol Cell Biol NG-A2

Title: Synaptic activation tunes action potential shape through a BK-channel-mediated feedback pathway

Authors: *E. S. NIKITIN[1], M. V. ROSHCHIN[1], M. MATLASHOV[2], V. N. IERUSALIMSKY[1], P. M. BALABAN[1], V. BELOUSOV[2], G. KEMENES[3], K. STARAS[3]
[1]Inst. Higher Nervous Activity, Moscow, Russian Federation; [2]Shemyakin-Ovchinnikov Inst. of Bioorganic Chem., Moscow, Russian Federation; [3]Sussex Neuroscience, Sch. of Life Sciences, Univ. of Sussex, Brighton, United Kingdom

Abstract: In central mammalian neurons, large conductance Ca2+-activated voltage-dependent K+ channels (BK channels) are known to influence the repolarization phase of action potentials and serve as a unique detector of coincidental intracellular Ca2+ elevation and membrane depolarization. This raises the possibility that the spike-evoked transient Ca2+ rises in synaptic boutons or axonal structures could potentially drive BK channel activity and thus contribute to shaping subsequent spikes, providing a novel feedback mechanism linking synaptic function and AP expression. Here we investigate this in layer 5 cortical neurons using Ca2+ uncaging and

imaging, patch clamp recording, and channel blockers. We show that a single spike-driven Ca2+ rise in a proximal synaptic bouton or first node of Ranvier significantly narrows the subsequent action potential and that this is mediated by a form of BK channel-dependent intrinsic feedback. This activity-dependent plasticity mechanism likely serves to limit runaway spike-broadening during repetitive firing, allowing a neuron to fine-tune its action potential expression based on synaptic firing history.

Disclosures: E.S. Nikitin: None. **M.V. Roshchin:** None. **M. Matlashov:** None. **V.N. Ierusalimsky:** None. **P.M. Balaban:** None. **V. Belousov:** None. **G. Kemenes:** None. **K. Staras:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.21/D60

Topic: B.04. Ion Channels

Title: Cav2.3 contributes to hippocampal excitability through activation of calcium-dependent potassium channels

Authors: *J. J. GUTZMANN, D. A. HOFFMAN
NICHD, NIH, Bethesda, MD

Abstract: Fragile X syndrome (FXS) is an inherited form of intellectual disability with a high co-morbidity with autism and epilepsy. FXS results from a mutation in a single gene, Fragile X Mental Retardation Protein (FMRP), on the X chromosome. FMRP is an mRNA regulating protein, thus its loss leads to the perturbation of a wide range of cellular functions and endows FXS with a challenging, multi-facetted disease presentation. One of the prominent features of FXS is neuronal hyperexcitability and propensity for epilepsy, suggesting the potential dysregulation of voltage-gated ion channels.
High-throughput sequencing of mRNAs regulated by FMRP, and thus potentially disrupted in FXS, has recently identified voltage-gated Ca^{2+} (Cav2.3) and K^{+} (Kv4.2) channels to be among the affected proteins in FXS. We investigated the effect of Cav2.3 loss on the electrophysiological properties and neuronal excitability in the mouse hippocampus, a brain area important for learning, and prone to epileptic activity. Using whole-cell patch-clamp recordings from hippocampal CA1 pyramidal neurons, we found that these cells exhibited higher firing frequencies, larger frequency-dependent action potential broadening, and reduced after-hyperpolarization in Cav2.3-KO mice compared to WT. Using pharmacological blockade of different channels involved in action potential generation, we show that the hyperexcitability is caused by a reduced function of Ca^{2+}-dependent K^{+} channels. Specifically, using whole-cell and single-channel recordings, we find that both Ca^{2+}-gated (BK) and Ca^{2+}-modulated (Kv4.2) K^{+}

currents are significantly reduced in Cav2.3-KO neurons.To elucidate how the altered membrane properties of CA1 neurons affect their output function, we analyzed the short-term plasticity between CA1 and the subiculum, a brain region downstream of CA1. Paired pulse facilitation experiments demonstrated a much larger facilitation effect in Cav2.3-KO animals compared to WT, in line with altered action potentials observed in CA1.In summary, our data shows for the first time that Cav2.3 is the Ca^{2+} source for Ca^{2+}-dependent K^{+} channels regulating action potential repolarization. This leads to increased excitability and altered synaptic plasticity. Cav2.3 emerges therefore as a promising target for addressing circuit level hyperexcitability in FXS mouse models.

Disclosures: J.J. Gutzmann: None. **D.A. Hoffman:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.22/D61

Topic: B.04. Ion Channels

Support: NIH Grant DC01919

NIH Grant NS18492

NIH Training Grant GM007324

Title: Activation of Slack potassium channels triggers an increase in mRNA translation

Authors: *T. J. MALONE[1], P. LICZNERSKI[2], E. A. JONAS[2], L. K. KACZMAREK[3]
[1]Cell. and Mol. Physiol., [2]Intrnl. Medicine/Section Endocrinol., [3]Cell. and Mol. Physiol./Pharmacology, Yale Univ. Sch. of Med., New Haven, CT

Abstract: The Slack ion channel is a member of a family of large conductance sodium-activated potassium channels. It is expressed predominantly in neurons of the central nervous system where it regulates neuronal excitability. FMRP, an important regulator of mRNA translation, binds both Slack mRNA and the Slack protein. The association of Slack with FMRP stimulates channel activity, raising the possibility that activation of Slack channels may also regulate the function of FMRP. Our laboratory has previously identified Slack as required for a protein translation-dependent recovery from an extended period of inhibition in Aplysia neurons following stimulation, further suggesting that Slack may play a role in the regulation of mRNA translation. Here we provide the initial evidence for such a role. We transfected cells with a fluorescent reporter for mRNA translation, which contains the 5’ and 3’ sequences of the mRNA for β-actin, but with the coding region replaced with that for the irreversibly-photoconvertible fluorescent protein dendra2. We were able to visualize real-time translation in mouse cortical

neurons and in HEK cell cultures. Based on the observed translation levels in neurons from wild-type, Slack knockout, and FMRP knockout mice, we propose a mechanism whereby Slack activation causes an increase in translation that is enhanced in the absence of FMRP. Additionally, experiments in HEK cell culture using silencing RNA knockdowns suggest that the Slack binding partners CYFIP1, another FMRP binding protein; and Phactr-1, a PP1 binding protein, also modulate this activation-dependent translation. This mechanism of Slack-dependent translation potentially represents the first instance of the direct modulation of mRNA translation by activation of an ion channel.

Disclosures: T.J. Malone: None. **P. Licznerski:** None. **E.A. Jonas:** None. **L.K. Kaczmarek:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.23/D62

Topic: B.04. Ion Channels

Title: Evidence for the participation of TASK potassium channels in formalin induced acute and chronic inflammatory pain

Authors: *R. NORIEGA, G. GARCÍA, V. A. MARTÍNEZ-ROJAS, J. MURBARTIÁN
Farmacobiologia, Cinvestav, Ciudad De Mexico, Mexico

Abstract: Inflammatory pain is generated as a consequence of tissue damage, where various inflammatory mediators such as H^+ are produced, which cause extracellular acidification and increase excitability. TASK potassium channels activation is inhibited by acidification of extracellular medium, which makes them potential candidates as regulators of nociceptive transmission. In this study the participation of TASK channel in acute and chronic inflammatory pain induced by formalin was determined. In the acute inflammatory pain model, subcutaneous formalin injection (1%, in PBS) on the dorsal surface of the right hind paw of Wistar rats, produced a characteristic biphasic response of the test. Pretreatment with ML365 specific blocker for TASK-1 (100μM-3mM) and PK-THPP for TASK-3 (3μM-300μM), administered intrathecally, significantly increased the effect of nociceptive responses in both phases, in a dose-dependent manner. Chronic inflammatory pain was assessed 6 days after injection of formalin into the right paw by testing secondary mechanical allodynia and hyperalgesia behaviors evoked by mechanical stimulation with the use of von Frey filaments. Pretreatment with the TASK1 and TASK3 channel blockers significantly increased allodynia in a dose-dependent manner and hyperalgesia only increased with PK-THPP at 100μM. In post-treatment the drugs significantly increase both behaviors, in a dose-dependent manner. The pharmacological and behavioral data suggest that blockade of TASK potassium channels produces a pro-allodynic and pro-

hyperalgesic effect, whereby TASK potassium channels play an important role in reducing acute inflammatory pain and reducing the development and maintenance of allodynia and hyperalgesia in chronic inflammatory pain.

Disclosures: R. Noriega: None. **G. García:** None. **V.A. Martínez-Rojas:** None. **J. Murbartián:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.24/D63

Topic: B.04. Ion Channels

Support: NIH NIDCD RO1 DC013080

NIH NIDCD F31 DC03665

NSF EAPSI Fellowship

NSF #1058957

FSU Legacy Fellowship

TMH Bryan Robinson Endowment

Title: Facile conjugation of the peptide margatoxin to luminescent quantum dots for targeted delivery towards the voltage-gated potassium channel Kv1.3 in the olfactory bulb

Authors: *A. SCHWARTZ[1], A. KAPUR[2], Z. HUANG[3], R. ANANGI[5], Z. DEKAN[5], E. FARDONE[3], G. PALUI[2], G. F. KING[5], H. MATTOUSSI[2], D. A. FADOOL[4]
[1]Florida State University, Mol. Biophysics Program, Tallahassee, FL; [2]Dept. of Chem., [3]Program in Neuroscience, Dept. of Biol. Sci., [4]Mol. Biophysics Program, Program in Neuroscience, Dept. of Biol. Sci., Florida State Univ., Tallahassee, FL; [5]Inst. of Mol. Biosci., The Univ. of Queensland, Brisbane, Australia

Abstract: Venom-derived ion channel inhibitors typically have high levels of channel selectivity, potency and stability, but, like most drug molecules, targeting to a specific location can be problematic. Kv1.3 is a voltage-gated potassium channel (K*v*) that has select distribution and is well characterized for its role in immunity, glucose metabolism and olfaction. Our interests lie in the ability of Kv1.3 to regulate excitability in mitral cells of the olfactory bulb (OB), where it has the potential to serve as a metabolic target to balance body weight and enhance olfactory ability. To aid in targeted delivery, we have developed protocols for conjugating the venom-derived Kv1.3 inhibitor margatoxin (MgTx) to luminescent quantum dots

(QDs). Previously, we described a protocol using carbodiimide crosslinking chemistry for effective conjugation of the native peptide to QDs and observed a retention of biophysical properties associated with block of the vestibule of Kv1.3 by the QD-MgTx conjugate compared to that of MgTx. Here, we have improved upon our initial methodology such that MgTx could be conjugated to QDs in a controlled orientation and facile manner through insertion of a polyhistidine tag on the N-terminus of MgTx (HisMgTx). Using recombinant expression and synthetic chemistry, we produced three forms of HisMgTx. When measured by whole-cell patch-clamp electrophysiology, the produced peptides potently inhibited 80% of the outward current in Kv1.3-transfected HEK-293 cells and had an IC_{50} of 100 - 350 pM. When conjugating to QDs using His-conjugation, as many as 40 HisMgTx peptides can be bound to an individual QD, as quantified by a shift in mobility on an agarose gel. This QDHisMgTx conjugate labeled Kv1.3 expressing HEK-293 cells with 90 nanograms of peptide. Using whole-cell, patch-clamp electrophysiology, the QDHisMgTx conjugate could similarly inhibit 80% of the outward current flow in Kv1.3 expressing HEK-293 cells. Towards targeted delivery of the QDHisMgTx conjugate to Kv1.3 in the OB, we have begun preliminary experiments using surgically-implanted osmotic mini-pumps and intranasal delivery approaches, where we are observing the effects of the conjugate on whole-body metabolism and its distribution following delivery.

Disclosures: **A. Schwartz:** None. **A. Kapur:** None. **Z. Huang:** None. **R. Anangi:** None. **Z. Dekan:** None. **E. Fardone:** None. **G. Palui:** None. **G.F. King:** None. **H. Mattoussi:** None. **D.A. Fadool:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.25/E1

Topic: B.04. Ion Channels

Support: AHA SDG Grant

National Ataxia Foundation Grant

Title: Structure-activity relationship studies of SK channel positive allosteric modulators

Authors: Y. NAM, R. ORFALI, A. VIEGAS, *M. ZHANG
Chapman Univ. Sch. of Pharm., Irvine, CA

Abstract: The cerebellar Purkinje Cells (PCs) are affected in many types of ataxia. The dysfunction of PCs, more specifically the loss of firing precision of PCs is one of the primary mechanism underlying the symptom of ataxia, including spinocerebellar ataxia. Disruptions of regular pacemaking activity of PCs have been identified in studies with mouse models of ataxia.

Drugs that normalize the regular firing of PCs have been suggested as therapeutics for the symptom of ataxia patients. Small conductance Ca^{2+} activated potassium (SK) channels emerged as one of the principle ion channels involved in PCs pacemaking. Positive allosteric modulators (PAMs) targeting the SK channels have been shown to alleviate behavioral and neuro-pathological symptom in animal models of ataxia. Despite this progress, many SK channel PAMs often suffer from low potency and/or poor pharmacokinetic properties. Lack of knowledge about the binding site for the compounds was a major reason that hinders the development of more effective therapeutics targeting SK channels. We recently determined the crystal structures of the PAMs such as 1-EBIO and NS309 in complex with their binding pocket. Further analyses show a strong correlation between the potency of the PAMs and their interaction energy within the binding pocket. These structure-activity relationship studies of SK channel modulation by PAMs might facilitate drug discovery targeting SK channels.

Disclosures: **Y. Nam:** None. **R. Orfali:** None. **A. Viegas:** None. **M. Zhang:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.26/E2

Topic: B.04. Ion Channels

Support: NHLBI R01-102758

NHLBI 5T32GM008181-28

Amer. Physiological Society Ryuji Ueno Award sponsored by the S&R Foundation

Title: The effects of single nucleotide polymorphisms (SNPs) on human BK channel currents

Authors: *A. PLANTE, *A. PLANTE, *A. PLANTE, B. MCNALLY, M. LAI, A. MEREDITH
Physiol. Dept., Univ. of Maryland, Baltimore, Baltimore, MD

Abstract: Large-conductance, Ca^{2+}-activated K^+ (BK) channels are important regulators of membrane excitability. One mechanism with the potential to alter BK currents is natural genetic variation via single nucleotide polymorphisms (SNPs). The BK channel gene, *Kcnma1*, contains >100 nonsynonymous SNPs, but the effects of most SNPs on BK function are unknown. Using standard patch clamp electrophysiology in HEK293 cells, we screened the effects of six SNPs (A138V, C495G, N599D, R800W, R640Q/R645Q) selected based on either disease linkage or their predicted ability to alter BK current properties. C495G and R800W had the largest impact on BK currents. In symmetrical and physiological K^+, C495G significantly shifted the half-maximal voltage of activation ($V_{1/2}$) to more hyperpolarized potentials (-15 to -20 mV) and

speeded activation, indicating C495G confers gain of function properties. In contrast, R800W significantly shifted the $V_{1/2}$ to more depolarized potentials (+15 to +35 mV) and slowed activation in symmetrical K^+, indicating R800W confers loss of function properties. Surprisingly, R800W slowed activation but did not shift the $V_{1/2}$ in physiological K^+. Although C495 was previously identified as a redox modulated residue, C495G did not alter the effects of peroxide or DTT on BK currents, suggesting C495G does not produce a gain of function effect by altering redox modulation of the BK channel. To explore which amino acid properties were responsible for the loss of function effects of R800W, R800 was substituted for residues with different (R800A, R800E, R800Q) or similar (R800F) properties to W. No substitution recapitulated R800W current properties, indicating that the altered size, charge, and hydrophobicity alone does not fully account for the loss of function effect of R800W. The impact of C495G and R800W on action potential (AP)-evoked BK currents was tested with AP commands from suprachiasmatic nucleus neurons, sinoatrial node, and bladder myocytes. C495G significantly increased the peak amplitude of AP-evoked BK currents, while R800W did not have an effect. However, when R800W was expressed in parallel with the epilepsy-linked, gain of function mutation D434G (D434G/R800W), the $V_{1/2}$ was not shifted, but activation was slowed and the peak amplitude of AP-evoked BK current was decreased compared to D434G alone. These results suggest that in a physiological context, C495G can have a gain of function effect, while R800W could oppose the aberrant effects of an epilepsy-linked mutation on the BK current properties. These results suggest natural genetic variation (SNPs) in the BK channel could potentially alter the severity and penetrance of D434G-linked epilepsy.

Disclosures: **A. Plante:** None. **B. McNally:** None. **M. Lai:** None. **A. Meredith:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.27/E3

Topic: B.04. Ion Channels

Support: NIG Grant 1R21NS097899

Title: Decreased expression of Kir K^+ channel in iPSC-derived astrocytes in subjects with Monge's disease

Authors: W. WU[1], *H. YAO[1], J. WANG[1], G. G. HADDAD[1,2,3]
[1]Dept Pediat, UCSD, LA Jolla, CA; [2]Dept Neurosci, UCSD, La Jolla, CA; [3]Rady Children's Hospital-San Diego, San Diego, CA

Abstract: Monge's disease, also called chronic mountain sickness (CMS) is a disease in highlanders whose blood has abnormal low level of oxygen. The prevalence of CMS patients can

be as high as 15-20% while the majority of highlanders (non-CMS) are rather healthy in high altitudes. CMS patients typically experience a variety of neurologic symptoms such as migraine, mental fatigue, confusion, dizziness, loss of appetite and memory loss. The cellular and molecular mechanisms underlying CMS neuropathology is not fully understood. Our previous study employed an induced pluripotent stem cells (iPSCs) approach and revealed that altered sodium channel properties and resulting decreased neuronal excitability in the iPSCs-derived neurons from CMS patients. Here, we further characterize the electrophysiological properties of iPSCs-derived astrocytes from both CMS and non-CMS subjects. Our patch clamp experiments showed that there was no significant differences of cell capacitance and input resistance between CMS and non-CMS astrocytes. The membrane potential of CMS group (-24.9±2.7 mV, n=44) is significantly more depolarized than that of non-CMS group (-43.9±3.7, n=26) (p<0.01). We further compared the current densities of each type of ion channels expressed in the iPSC-derived astrocytes. We found that the current densities of the inwardly-rectifying potassium (Kir) channels in CMS astrocytes (-5.7±2.2 pA/pF at -140 mV, n=20) were significantly reduced as compared to non-CMS subjects (-28.4±3.4 pA/pF at -140 mV, n=30) (p< 0.01). In contrast, there were no significant differences of current densities of voltage-gated potassium channels between CMS and non-CMS astrocytes. We also found a subpopulation (~15%) of astrocytes expressing voltage-gated sodium channels and the current density in CMS astrocyte was not altered. Finally, inward currents induced by stepping extracellular [K^+] from 3 to 12 mM and 60 mM (reflecting K^+ uptake ability) in CMS astrocytes (-12.7±6.4 pA and -290.8±182.0 pA) is significantly less than that in non-CMS astrocytes (-341.6±210.6 pA and -1484.3±374.9pA) (n=5-17, p<0.05). Since glial Kir K^+ channels are critical for K^+ homeostasis and regulation of neuronal excitability in brain, our findings of decreased Kir channels and K^+ uptake ability in CMS astrocytes indicate a potential likely link between CMS symptoms and the Kir channel abnormalities.

Disclosures: **W. Wu:** None. **H. Yao:** None. **J. Wang:** None. **G.G. Haddad:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.28/E4

Topic: B.04. Ion Channels

Support: CIHR

NSERC

Title: Differential regulation of delayed-rectifying voltage-gated potassium channel dynamics by the mutant sigma1-receptor underlying ALS16

Authors: *M. J. ABRAHAM[1], A. Y. WONG[2], R. BERGERON[2,1]
[1]Dept. of Cell. and Mol. Medicine, Neurosci. Program, Univ. of Ottawa, Ottawa, ON, Canada; [2]Ottawa Hosp. Res. Inst., Ottawa, ON, Canada

Abstract: The Sigma-1 Receptor (sig-1R) is a highly conserved endoplasmic reticulum (ER) resident protein, which is expressed in most cell types throughout the body. It involved in diverse functions, including modulation of plasma-membrane ion channels, mediation of Ca^{2+} signalling from the ER to the mitochondria, and the ER stress response. Unexpectedly for a protein with such ubiquitous expression, mutations in the sig-1R have been associated with neuromuscular disease. The most widely described mutation is a single point mutation, E102Q, which results in a severe, juvenile-onset form of Amyotrophic Lateral Sclerosis (ALS16). As a hallmark of ALS is motorneuron hyperexcitability and the sig-1R modulates ion channels that control action potential repolarization, we speculated that aberrant modulation of these channels by sig-1R-E102Q may represent a hitherto uncharacterized mechanism of neuronal excitability in ALS. To test this, we transiently transfected HEK293 cells with a neuronal (Kv1.2) or a cardiac (Kv1.5) delayed rectifier K^+-channel subunit together with either WT sig-1R or sig-1R-E102Q. Electrophysiological recording of whole cell K^+ currents from these cells show that activation of the sig-1R results in a 20% decrease in current amplitude for both Kv1.2 and Kv1.5. Both Kv1.2 and Kv1.5 show a significant increase in steady-state inactivation with increasing voltage, and Kv1.2 inactivation was significantly enhanced following sig-1R activation. When HEK293 cells were transfected with sig-1R-E102Q, the voltage-dependence of steady-state inactivation of Kv1.2 was abolished, voltage-dependence of inactivation was inhibited, yet activation of sig-1R-E102Q produced no additional modulation. In contrast, steady-state inactivation and voltage-dependence of inactivation of Kv1.5 was less affected by sig-1R-E102Q Despite their high homology, our data suggest that there is differential modulation of Kv1.2 and Kv1.5 by the sig-1R. Furthermore, we observe aberrant modulation of only neuronal Kv channels (Kv1.2) by sig-1R-E102Q, which may underlie the hyperexcitability phenotype in ALS. Taken together, our work provides insights into why sig-1R mutations only affect motorneurons but leave all other systems apparently intact.

Disclosures: **M.J. Abraham:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Canadian Institute of Health Research. **A.Y. Wong:** None. **R. Bergeron:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Canadian Institute of Health Research.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.29/E5

Topic: B.04. Ion Channels

Support: Canadian Institutes for Health Research

Canada Foundation for Innovation

Title: Phosphorylation-specific interaction of potassium channels with Fragile X mental retardation protein tunes inhibitory neurotransmission

Authors: *Y. YANG[1,2], J. ARSENAULT[2,4], A. BAH[3], M. KRZEMINSKI[3], A. FEKETE[2,4], L. PACEY[5], A. WANG[2,4], J. FORMAN-KAY[3,6], D. HAMPSON[5,7], L.-Y. WANG[2,4]
[1]Univ. of Minnesota Med. Sch. Duluth Campus, Univ. of Minnesota, Duluth, MN;
[2]Neurosciences & Mental Hlth., [3]Mol. Med., SickKids Res. Inst., Toronto, ON, Canada;
[4]Physiol., [5]Pharmaceut. Sci., [6]Biochem., [7]Pharmacol., Univ. of Toronto, Toronto, ON, Canada

Abstract: Principal neurons encode and project information by varying their firing rates and patterns that are precisely defined by inhibition from GABAergic interneurons. However, the molecular basis underlying inhibitory control remains elusive. We find that excessive presynaptic GABA release from interneurons dramatically attenuates the firing rate of Purkinje neurons in the cerebellum of Fragile X mental retardation protein (FMRP) knockout mice. This inhibitory overtone is resulted from increased excitability of the interneuron terminals where Kv1.2 potassium channels are downregulated in the absence of FMRP. We further reveal that the N-terminus of FMRP directly binds the C-terminus of Kv1.2, only when specific serine residues in Kv1.2 are phosphorylated. This interaction promotes the function of Kv1.2 at the nerve terminal, providing a key mechanism for dynamic tuning of inhibition; and further understanding of the novel molecular locus will help develop genetic and pharmacological therapies to rectify excitation/inhibition imbalance in neuropsychiatric disorders such as Fragile X syndrome and autism.

Disclosures: **Y. Yang:** None. **J. Arsenault:** None. **A. Bah:** None. **M. Krzeminski:** None. **A. Fekete:** None. **L. Pacey:** None. **A. Wang:** None. **J. Forman-Kay:** None. **D. Hampson:** None. **L. Wang:** None.

Poster

375. Potassium Channels I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 375.30/E6

Topic: B.04. Ion Channels

Support: NIH/NIDA Intramural Research Program

Title: Expression of potassium channels in compulsive methamphetamine-seeking & abstinent rats using a punishment model

Authors: *S. JAYANTHI[1], L. CONTU[2], B. LADENHEIM[2], M. T. MCCOY[2], M. JOB[2], B. CAMPBELL[2], C. BLACKWOOD[2], J. CADET[2]
[1]Mol. Neuropsychiatry Br., [2]Mol. Neuropsychiatry Res. Br., NIDA-IRP, Baltimore, MD

Abstract: Methamphetamine (METH) is a highly addictive stimulant. METH addicts continue to take the drug despite adverse medical, social, financial, and legal consequences. To investigate the biochemical and molecular dysfunctions that may subsume METH addiction we used a self-administration model with footshock paradigm in the present study. Foot-shocks cause animals to divide into a subpopulation of rats that continue to lever press for METH even in the presence of footshocks: these rats are called shock-resistant (SR) or “addicted” rats. In contrast, other rats progressively decreased their METH intake with increasing shock intensity. These are shock-sensitive (SS) or “non-addicted” rats. In addition, we also included two additional controls groups that were yoked to receive non-contingent footshocks when SR (yoked SR) and SS (yoked SS) rats received footshocks. Herein, we have measured the expression of genes that code for several subfamilies of potassium channels. Interestingly, the SR rats showed increased mRNA expression of *inward-rectifier* potassium channels (Kir-*Kir2.1 and Kir2.4*) in comparison to controls, YSR, and SS groups. In contrast, the SS rats showed increased mRNA expression of *Kir1.1* in comparison to control and YSS rats. Together, our results provide further evidence for the potential involvement of potassium channels in METH addiction.
Acknowledgement: This work is supported Department of Health and Human Services/ National Institutes of Health/ National Institute on Drug Abuse/ Intramural Research Program.

Disclosures: S. Jayanthi: None. **L. Contu:** None. **B. Ladenheim:** None. **M.T. McCoy:** None. **M. Job:** None. **B. Campbell:** None. **C. Blackwood:** None. **J. Cadet:** None.

Poster

376. Potassium Channels II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 376.01/E7

Topic: B.04. Ion Channels

Support: NIH R01HD072056

NIH R01NS044163

Title: Phosphatidylinositol 4,5-bisphosphate (PIP_2) regulates afterhyperpolarizations by modulating voltage-gated Ca^{2+} channels in oxytocin neurons of the supraoptic nucleus

Authors: *M. KIRCHNER[1], R. C. FOEHRING[3], W. E. ARMSTRONG[2]
[2]Neurosci. Inst., [1]Univ. of Tennessee Hlth. Sci. Ctr., Memphis, TN; [3]Anat. & Neurobio., Univ. Tennessee Hlth. Sci. Ctr., Memphis, TN

Abstract: Oxytocin- (OT) and vasopressin- (VP) secreting magnocellular neurosecretory cells play a crucial role in numerous physiological functions including lactation, parturition (OT) and cardiovascular regulation (VP). The release of both hormones is optimized by a burst-firing pattern of action potentials. The after-hyperpolarization (AHP) plays a crucial role in shaping bursts and thus is critical for robust hormone release. Previous work in our lab demonstrated that availability of the membrane-bound lipid phosphatidylinositol 4,5-bisphosphate (PIP_2) modulates the medium (mAHP) and slow (sAHP) duration AHPs in supraoptic neurons. Using simultaneous whole cell recording, calcium imaging, and photolytic uncaging, we evaluated the interaction of PIP_2 availability and specific Ca^{2+} channels with AHPs of both OT and VP cell types in brain slices. We previously showed that depleting PIP_2 blocked both mAHP & sAHP, as well as inhibited somatic Ca^{2+} in OT neurons while leaving these measures unaffected in VP neurons. This hinted that PIP_2 was inhibiting Ca^{2+} entry in OT neurons. This idea was supported when AHPs were generated by photolytic uncaging of Ca^{2+}, where we observed no inhibition of either the mAHP or sAHP by PIP_2 depletion. Furthermore, PIP_2 depletion significantly inhibited isolated whole cell Ca^{2+} currents in OT neurons, demonstrating that PIP_2 controls Ca^{2+} entry. Previous work has shown that coupling of specific voltage-gated Ca^{2+} channels (VGCCs) to the AHP is cell type dependent. Therefore, we tested the VGCC blockers 5 µM nifedipine (L-type), 1 µM conotoxin GVIA (N-Type), and 500 nM agatoxin-IVA (P/Q-type) on the mAHP & sAHP. Only conotoxin GVIA had a significant effect on the AHP, inhibiting the mAHP and sAHP components by 65% and 40%, and corresponding peak $[Ca^{2+}]_i$ by 61% & 73%, respectively in OT neurons. VP neurons displayed smaller inhibition of mAHP & sAHP (25% & 26%) while demonstrating similar $[Ca^{2+}]_i$ reductions to OT neurons (59% & 71%). Small reductions of $[Ca^{2+}]_i$ were observed in response to nifedipine and agatoxin-IVA, even though both AHPs were unaffected. Application of Cd^{2+} inhibited the mAHP by 90% and abolished the sAHP at 1000 ms

in voltage clamp, suggesting that a strong majority of the current is VGCC-dependent, but that a Ca^{2+}-independent component exists. Future experiments will be designed to verify N-channel modulation by PIP_2, and determine the mechanism of this modulation in OT neurons.

Disclosures: M. Kirchner: None. **R.C. Foehring:** None. **W.E. Armstrong:** None.

Poster

376. Potassium Channels II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 376.02/E8

Topic: B.04. Ion Channels

Support: NIH Grant NS078184

Title: Regulation of sodium-dependent potassium channel slack by 14-3-3 zeta (YWHAZ) in dorsal root ganglion neurons

Authors: *R. G. POWELL[1], K. D. PRYCE[2], S. GURURAJ[3], A. BHATTACHARJEE[3]
[1]SUNY Univ. At Buffalo, Buffalo, NY; [2]Pharmacol., State Univ. of New York At Buffalo, Buffalo, NY; [3]Univ. at Buffalo - The State Univ. of New York, Buffalo, NY

Abstract: Tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation proteins (14-3-3), also known as PKC inhibitory proteins (KCIP-1), are a family of adaptor proteins that bind to specific recognition motifs denoted as: mode-1 (RSXpS/TXP) and mode-2 (RXXXpSXP). The 14-3-3 family of proteins are ubiquitously expressed in the central nervous system where they mediate phospho-regulatory pathways including: protein trafficking, signal transduction, metabolism, apoptosis, cell proliferation, and cycle regulation. In this study, we investigated the role of 14-3-3 zeta in Protein Kinase C (PKC) regulation of Slack channels. The Slack C-terminus contains several predicted mode-1 14-3-3 binding sites. Using proteomic mass spectrometry we determined that 14-3-3 zeta belongs to the Slack channel interactome in dorsal root ganglion (DRG) neurons. Next, we conducted co-immunoprecipitation assays and immunohistochemistry to confirm that Slack channels and 14-3-3 zeta co-localize and bind with each other in intact DRG neurons and within the dorsal horn of the spinal cord. Secondly, to determine the functional role of the Slack/14-3-3 interaction, we used whole-cell patch-clamp to measure changes heterologous expressed Slack currents. We found that co-expression of 14-3-3 zeta did not affect the Slack current, however, in the presence of the PKC activator PMA, 14-3-3 zeta was found to inhibit the typical PMA induced up-regulation of Slack-current. Using biotinylation assays we demonstrated that after PMA stimulation there was a robust increase in membrane Slack expression, whereas membrane Slack expression decreased in cells co-transfected with Slack and 14-3-3 zeta after stimulation by PMA. Finally, we investigated the regulation of 14-3-3 zeta on Slack channels in native neurons. We used small interfering RNAs

to knockdown 14-3-3 zeta expression. siRNA mediated knockdown of 14-3-3 zeta resulted in an unexpected decrease total potassium current, broadening of action potential duration and repetitive firing when compared to DRG neurons treated with scrambled siRNA. Finally, in behavioral studies 14-3-3 zeta knockout mice exhibit a pronounced basal thermal hyperalgesia. Overall, our data strongly suggests 14-3-3 zeta is a powerful regulator of Slack currents and DRG neuronal excitability.

Disclosures: R.G. Powell: None. **K.D. Pryce:** None. **S. Gururaj:** None. **A. Bhattacharjee:** None.

Poster

376. Potassium Channels II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 376.03/E9

Topic: B.04. Ion Channels

Support: NIH Grant NS078184

Title: PKA-induced Slack channel internalization from the neuronal membrane occurs by AP-2 Clathrin mediated endocytosis

Authors: *A. BHATTACHARJEE[1], S. GURURAJ[1], K. EVELY[2], K. D. PRYCE[3]
[1]Univ. at Buffalo - The State Univ. of New York, Buffalo, NY; [2]Univ. At Buffalo - Downtown Campus, Buffalo, NY; [3]Pharmacol., State Univ. of New York At Buffalo, Buffalo, NY

Abstract: The sodium-activated potassium (K_{Na}) channel Slack (Kcnt1) is abundantly expressed in nociceptor neurons of the dorsal root ganglion (DRG), where they transmit the large outward conductance I_{KNa} and arbitrate membrane excitability. Expression of Slack channels at the DRG membrane is necessary for their characteristic firing accommodation during maintained stimulation and reduced membrane channel density produces hyperexcitability (Nuwer et al., J Neurosci 30:14165-72, 2010). We have previously shown that in the pro-inflammatory state, decrease in channel density leading to reduced Slack-mediated I_{KNa} underlies DRG neuronal sensitization. An important component of inflammatory signaling, Protein Kinase A (PKA) caused Slack channels to internalize from the DRG membrane, reduced I_{KNa} and produced DRG hyperexcitability when activated in cultured primary DRG neurons. Here, we show that this PKA-induced retrograde trafficking of Slack channels occurs *ex vivo* in intact spinal cord slices and that it is carried out by AP-2 clathrin-mediated endocytosis (CME) in DRG neurons. We provide mass spectrometric and biochemical evidence of native neuronal AP-2 adaptor proteins' association with Slack channels and specifically, that the binding site is a di-leucine motif housed in the cytoplasmic Slack C-terminus. We created a competitive peptide blocker of AP-2-Slack binding to empirically demonstrate that this interaction is essential for Clathrin recruitment

to the DRG membrane, Slack channel endocytosis and resultant DRG hyperexcitability, subsequent to PKA activation. Together, these findings identify AP-2 and Clathrin proteins as novel players in Slack channel regulation. Given the significant role played by Slack channels in nociceptive neuronal excitability, the AP-2 CME trafficking mechanism provides a promising approach to target peripheral and possibly, central neuronal sensitization.

Disclosures: **A. Bhattacharjee:** None. **S. Gururaj:** None. **K. Evely:** None. **K.D. Pryce:** None.

Poster

376. Potassium Channels II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 376.04/E10

Topic: B.04. Ion Channels

Support: NHMRC Grant 10915693

Title: Characterization of a novel mouse model of KCNT1 epileptic encephalopathy

Authors: *L. E. BURBANO[1,2], M. LI[1], N. JACOVSKI[1], K. L.-A. RICHARDS[1], E. GAZINA[1], S. MALJEVIC[1], C. REID[1,2], S. PETROU[1,2]
[1]Florey Inst. of Neurosci. and Mental Hlth., Melbourne, Australia; [2]Univ. of Melbourne, Melbourne, Australia

Abstract: Epilepsy of infancy with migrating focal seizures (EIMFS) is a severe form of childhood epilepsy characterized by an early onset, refractory seizures, global developmental delay and cognitive disability. *De novo* mutations of the sodium activated potassium channel gene KCTN1 have been found in up to 50% of the patients. The p.Pro924Leu mutation has been found in 2 patients with EIMFS and *in vitro* studies in heterologous expression systems have shown a gain of function effect of the mutation, resulting in large potassium currents compared to the wild type (WT) channel. We have characterized a knock-in mouse model expressing this gain-of-function mutation to explore the mechanisms by which the altered channel translates to the disease phenotype. To assess seizure susceptibility, video electrocorticogram(video/ECoG) analysis, thermal and chemoconvulsant (Pentylenetetrazole and Loxapine) tests were performed. Heterozygous Kcnt1$^{(L/+)}$ mice display no increased susceptibility to thermal or chemically induced seizures compared to Kcnt1 WT$^{(+/+)}$ mice. No differences were found on the video/ECoG for Kcnt1$^{(L/+)}$ and Kcnt1 WT$^{(+/+)}$ mice. In contrast, homozygous Kcnt1$^{(L/L)}$ mice exhibit spontaneous tonic clonic seizures, impaired nesting behaviour and a shortened lifespan. We also detected ictal discharges that coincide with convulsive seizures as well as interictal spikes in these mice. Kcnt1$^{(L/L)}$ mice provide a model of epileptic encephalopathy that will be valuable for studying the in vivo effects of gain of function KCNT1 mutations and the response to targeted therapeutic interventions.

Disclosures: **L.E. Burbano:** None. **M. Li:** None. **N. Jacovski:** None. **K.L. Richards:** None. **E. Gazina:** None. **S. Maljevic:** None. **C. Reid:** None. **S. Petrou:** None.

Poster

376. Potassium Channels II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 376.05/E11

Topic: B.04. Ion Channels

Support: Dean's Transformational Science Award

Sigma Xi Grant-in-Aid of Research

NIH Grant R21

Farber Family Foundation Award

Title: Inhibition of Kv3.4 channels is implicated in the potentiation of excitatory synaptic transmission in superficial dorsal horn neurons

Authors: *T. MUQEEM[1], V. PINTO[2], B. CHARSAR[1], E. BROWN[1], A. C. LEPORE[1], M. COVARRUBIAS[1]
[1]Dept. of Neurosci., Thomas Jefferson Univ., Philadelphia, PA; [2]ICVS/3Bs, Univ. of Minho, Braga, Portugal

Abstract: Kv3.4 channels underlie a majority of the high-voltage activating K^+ current in dorsal root ganglion (DRG) nociceptors, and are expressed in all functional compartments of these neurons, including synaptic terminals. Kv3.4 channels in DRG neurons regulate the rate of action potential repolarization in a manner that depends on phosphorylation of the channels' inactivation domain. Additionally, loss of Kv3.4 channel surface expression is implicated in the pathophysiology of spinal cord injury-induced chronic pain. We hypothesize that, through their ability to facilitate action potential repolarization, Kv3.4 channels help keep nociceptive synaptic transmission in check. Inhibition of the Kv3.4 channel would, therefore, promote pain transduction. To test this hypothesis, we have investigated excitatory synaptic transmission in the superficial dorsal horn under conditions that inhibit the Kv3.4 current in DRG nerve terminals. We used an ex vivo rat cervical spinal cord preparation suitable for whole-cell patch clamping of secondary neurons in the dorsal horn. Since Kv3.4 channels are hypersensitive to submillimolar concentrations of 4-aminopyridine (4-AP) and tetraethylammonium (TEA), we tested their effects on excitatory post-synaptic currents (EPSCs) in laminae I and II of the dorsal horn. We found that 500 μM TEA potentiates the EPSCs by 26% ($p = 0.01$, paired t-test, N=9). Similarly, 50 μM 4-AP potentiates the EPSCs by 34% ($p = 0.007$, paired t-test, N=9). Alternatively, the BK channel blocker iberiotoxin, the Kv7 channel blocker XE991, and the Kv1 channel blocker

dendrotoxin had little effect on the EPSCs. Paired pulse and spontaneous neurotransmission experiments were conducted to localize the effect to the presynaptic nerve terminal. Therefore, we conclude that the Kv3.4 channel expressed in nociceptors is a plausible regulator of nociceptive synaptic transmission in the spinal cord. Currently, we are exploring more specific knock down strategies to assess the role of the Kv3.4 channel as a regulator of nociception in vivo. Supported by grants from NIH, Farber Family Foundation, Sigma Xi Research Society and the Dean's Transformational Science Award from the Sidney Kimmel Medical College

Disclosures: T. Muqeem: None. **V. Pinto:** None. **B. Charsar:** None. **E. Brown:** None. **A.C. Lepore:** None. **M. Covarrubias:** None.

Poster

376. Potassium Channels II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 376.06/E12

Topic: B.04. Ion Channels

Support: NIH Grant DC012063

Title: KCNQ channels regulate firing properties of the calyx of Held

Authors: Y. ZHANG[1], L. O. TRUSSELL[2], *H. HUANG[1]
[1]Dept. of Cell and Mol. Biol., Tulane Univ., New Orleans, LA; [2]Oregon Hearing Res. Ctr., Oregon Hlth. and Sci. Univ., Portland, OR

Abstract: Globular bushy cells of the cochlear nucleus faithfully relay auditory signals to the medial nucleus of the trapezoid body (MNTB) principal neurons through a giant glutamatergic synapse, the calyx of Held. Reliability and precision of synaptic transmission are required in the calyx-MNTB synapse in order to encode timing with submillisecond delays at high frequencies. The waveform of the presynaptic action potential (AP) shapes the presynaptic calcium transient, and thus is crucial in determining timing and strength of synaptic transmission. During moderate to high-frequency trains, the presynaptic AP of several central synapses such as mossy fiber boutons are altered and the half-duration of action potentials is substantially increased. However, the calyx of Held shows very minor AP broadening at frequencies up to hundreds of hertz. We previously demonstrated that KCNQ5 channels control the presynaptic resting properties and release probability of the calyceal synapses. Here we found that KCNQ channels are activated during high-frequency AP activity, have profound effects on AP firing, and indeed are required to maintain the normal AP waveform of the calyceal terminal. Blocking KCNQ channels with XE991 increased presynaptic excitability, reduced the AP height, and broadened the AP duration during 333-Hz stimulation. Further experiments showed that blocking KCNQ leads to inactivation of presynaptic Na^+ and Kv1 channels and thereby indirectly affects the AP

waveform. Inactivation of Na^+ channels shortens the AP amplitude while inactivation of Kv1 broadens the AP and increases excitability. Application of low doses of TTX and margatoxin to partially block Na^+ and Kv1 channels could mimic the AP waveform change during high frequency stimulation. Resistance to changes in the AP waveform during activity ensures that the calyx can reproducibly trigger calcium influx and glutamate release. We conclude that KCNQ is activated during high-frequency activity and its activation prevents inactivation of Na^+ and Kv1 channels and thus plays key roles in maintaining a stable presynaptic AP waveform.

Disclosures: Y. Zhang: None. **L.O. Trussell:** None. **H. Huang:** None.

Poster

376. Potassium Channels II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 376.07/F1

Topic: B.04. Ion Channels

Support: NIH Grant HL112919

Title: KCNQ/K_V7 potassium channels contribute to the control of mouse nodose airway C-fiber excitability

Authors: *H. SUN, S. MEEKER, B. J. UNDEM
Johns Hopkins Univ., Baltimore, MD

Abstract: The M-current is carried by members of K_V7 family of K^+ channels encoded by *KCNQ* genes. It is now well established that the M-current is modulated by numerous mediators and functions as a "brake" on repetitive action potential (AP) discharges in both central and peripheral nervous systems. It has been shown that pharmacological activation of M-current suppresses epilepsy and alleviates pain. Yet, whether and how the KCNQ/K_V7 channel regulates the excitability of vagal airway nociceptive C-fibers remain to be elucidated. In this study we have used perforated patch clamp technique and extracellular recording of single Vagal nerve fiber terminal activities to address this issue. A voltage step from the holding potential of -30 mV to -60 mV evoked a small outward tail current and stepping back to -30 mV elicited a slowly activating and non-inactivating outward current in isolated mouse nodose neurons, both of which were nearly abolished by 10 μM XE991, a KCNQ/K_V7 channel blocker. The XE991-sensitive current was observed in every neuron studied. It activates around -65 mV with 50% activation being occurred at -38.2±2.1 mV and a maximal conductance of 10.3±1.3 nS/pF (n=8). The current has an activation constant of 212±23 ms at -30 mV and deactivates biexponentially. The KCNQ channel opener retigabine at 10 μM significantly accelerated its activation and slow down the deactivation, and negatively shifted its activation by 27 mV. Thus, the XE991-sensitive current recorded in mouse nodose neurons exhibits similar biophysical and pharmacological

properties previously reported for endogenous M-currents. Inhibition of M-current by XE991 slightly but significantly depolarized the nodose neurons (-60.1±1.2 vs. -62.9±1.0 mV, n=14) while opening the channels by retigabine markedly hyperpolarized the membrane (-73.1±0.9 vs. 60.4±0.7 mV, n=13) and this effect was reversed by simultaneous application of XE991. Retigabine also significantly increased the amount of depolarizing current needed to evoke an AP (25±5 *vs.* 133±20 pA, n=5). In a vagus-innervated isolated moue lung preparation, retigabine eliminated the spontaneous AP discharges in nodose C-fibers innervating the mouse lungs (n=3) and abolished AP discharge evoked by 3μM α,β-methylene ATP (AP number: 47 ± 7 vs. 0 ± 0, n=5). In conclusion, our results indicate that KCNQ/K_V7 channels play a role in maintaining the resting membrane potential of mouse nodose neurons and in regulating the excitability of vagal C-fibers at both the cell soma and nerve terminals within the airways. Activation of M-currents may represent a useful approach to suppressing airways hyperreflexivity associated with chronic cough, COPD, and asthma.

Disclosures: H. Sun: None. **S. Meeker:** None. **B.J. Undem:** None.

Poster

376. Potassium Channels II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 376.08/F2

Topic: B.04. Ion Channels

Support: DoD CDMRP grant W81XWH-15-1-0284

National Center for Advancing Translational Sciences

NIH Grant TL1 TR001119

DoD CDMRP grant W81XWH-15-1-0283

Title: Novel drugs that augment KCNQ (Kv7,"M-type") potassium channels as a post-event treatment for Traumatic Brain Injury

Authors: *M. HOBBS[1], F. A. BORGES-VIGIL[1], E. BOZDEMIR[2], R. J. VERAZA[1], V. BUGAY[1], L. ESPINOZA[1], D. M. HOLSTEIN[2], S. M. SPRAGUE[3], I. SANCHEZ[1], J. CARVAZOS[4], S. H. CHUN[2], R. BRENNER[1], J. LECHLEITER[2], M. S. SHAPIRO[1]
[1]Cell. and Integrative Physiol., [2]Cell and Structural Biol., [3]Neurosurg., [4]Neurol., UT Hlth. Sci. Ctr. SA, San Antonio, TX

Abstract: Traumatic brain injury (TBI) is a strong risk factor for the development of different neuronal disorders, such as epilepsy. Here, we hypothesize that reducing neuronal excitability early in the process of epileptogenesis moderates or prevents development of epilepsy. Since

enhancing KCNQ channel activity causes silencing of neurons and prevents seizures, we tested whether a drug that augments KCNQ K^+ currents, such as the FDA-approved antiepileptic drug, Retigabine (RTG), reduces development of seizures, cytotoxic brain cell swelling and the inflammatory response. We tested our hypothesis using the controlled closed-cortical impact TBI model, which mimics TBIs commonly experienced by civilians. Mice were administered RTG *(i.p. 1 mg/kg*) or vehicle within 30 minutes of TBI. Twenty-four hours after the TBI, EEG electrodes were implanted and electrical activity recorded the following day to assess spontaneous seizures. To assay for the reduction of seizure susceptibility by RTG treatment after TBIs, animals were challenged with pilocarpine (3X, 75 mg/kg, 6 days after TBI), and the percentage of animals responding with seizures compared between groups. Seizure susceptibility was assayed for 24 hours after pilocarpine; where the latency of the first seizure occurrence and percentage of animals that displayed seizures were both quantified. A different cohort of mice were sacrificed 24h after TBI and brain slices Nissl-stained to quantify cell soma areas. Finally, a third dual cohort of animals were sacrificed 6 days after CCCI and both hemispheres used for immunoblotting analyses of CD40L, a known marker of inflammation. We found that TBI caused spontaneous seizures in ~1/3 of vehicle only-treated mice. On the other hand, for animals treated with RTG, no animals displayed spontaneous seizures. Whereas 54% of the control animals displayed seizures within 24 hours after pilocarpine challenge, only 30% of RTG treated mice responded with seizures. RTG treatment also increased the latency to first seizure by about two-fold. RTG treatment also significantly reduced TBI-induced cytotoxic cell soma swelling 24h after TBI, and impaired TBI-induced increase in CD40L. Therefore, KCNQ-enhancing drugs may serve as a novel, effective treatment for the deleterious effects that follow TBIs.
Supported by the DoD CDMRP grants W81XWH-15-1-0284 (M.S, and R.B), W81XWH-15-1-0283 (J.L.), and by the National Center for Advancing Transnational Sciences, National Institutes of Health, through Grant TL1 TR001119.

Disclosures: M. Hobbs: None. **F.A. Borges-Vigil:** None. **E. Bozdemir:** None. **R.J. Veraza:** None. **V. Bugay:** None. **L. Espinoza:** None. **D.M. Holstein:** None. **S.M. Sprague:** None. **I. Sanchez:** None. **J. Carvazos:** None. **S.H. Chun:** None. **R. Brenner:** None. **J. Lechleiter:** None. **M.S. Shapiro:** None.

Poster

376. Potassium Channels II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 376.09/F3

Topic: B.04. Ion Channels

Support: NIH Grant NS073981

NIH Grant MH110887

Title: Ablation of KCNQ2/3 from either pyramidal neurons or interneurons leads to increased excitatory transmission in mouse hippocampus

Authors: H. SOH[1], K. SPRINGER[2], *A. TZINGOUNIS[3]
[1]Physiol. and Neurobio., [2]Univ. of Connecticut, Storrs, Storrs, CT; [3]Univ. of Connecticut, Storrs, CT

Abstract: In contrast to the wealth of knowledge on the role of KCNQ2/3 channels in excitatory neurons, the KCNQ2/3's ability to regulate interneuron excitability has been largely overlooked. To examine whether KCNQ2/3 channels could regulate interneuron intrinsic excitability, we developed mouse lines that lack *Kcnq2/3* specifically in MGE-derived PV^+ and SST^+ interneurons via Cre recombinase technology (using *Nkx2-1BAC*^{cre}; *SST-Cre;* or *PV-Cre* mice). We also crossed these mice to a reporter line (*Ai14*) that expresses the fluorescent protein tdTomato in cells in which Cre recombinase has been active, allowing for the identification of Cre-expressing PV^+ and SST^+ interneurons. Using these mice we have found that neocortical and hippocampal PV^+, but not SST^+, interneurons are more excitable in the absence of KCNQ2/3 channels. Surprisingly, we also found that ablation of *Kcnq2/3* from PV^+/SST^+ interneurons from an early period led to a significant elevation of the spontaneous and mini EPSC frequency (mEPSC) in CA1 pyramidal neurons later in development, at P16-P19. Importantly, these effects are specific to *Kcnq2/3* ablation from PV^+/SST^+ interneurons, as we did not find any changes to the mEPSC frequency in CA1 pyramidal neurons from *Kcnq2* pyramidal null neurons (*Emx1-Cre; Kcnq2*$^{f/f}$). Rather, in these neurons, the spontaneous EPSC frequency was elevated, consistent with increased axonal excitability. Therefore, KCNQ2/3 loss of function in either pyramidal neurons or interneurons could lead to network hyperexcitability.

Disclosures: H. Soh: None. **K. Springer:** None. **A. Tzingounis:** None.

Poster

376. Potassium Channels II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 376.10/F4

Topic: B.04. Ion Channels

Title: Suppressive effects of amyloid peptide fragments (1-42) and the toxic "core" (25-35) on Kv1.1 channel activity

Authors: *K. DEBOEUF, M. ISLAM, N. THELEN, J. FARLEY
Psychology and Neurosci., Indiana Univ. Bloomington, Bloomington, IN

Abstract: Past studies have shown that Aβ's role in the pathogenesis of Alzheimer's disease (AD) involves eventual disruption of Ca^{2+} homeostasis and synaptic communication, as well as

impairment of long-term potentiation (LTP), but the underlying mechanism(s) is still unclear. Work in the Farley lab suggests that Aβ-inhibition of voltage-dependent K^+ channels (e.g., Kv1.1) activity is among the earliest steps. Using murine Kv1.1 channels expressed in *Xenopus* oocytes, our lab previously elucidated a pathway in which Ca^{2+}-dependent activation of PP2B, PKC, PTKs, and RhoA all participated to produce rapid strong suppression of Kv1.1 activity. Because Kv1.1 and related channels are activated during an action potential, regulate depolarization Ca^{2+} influx, and inhibition of Kv1 channels can be neurotoxic, we speculate that Aβ-suppression of Kv1 channels could contribute to AD pathology. We assessed the effects of the Aβ(1-42) peptide and the core fragment [Aβ(25-35)] on Kv1.1 channels expressed in oocytes. Bath application of 1 μM Aβ(1-42) produced ~50% suppression of macroscopic Kv1.1 current within 30 m. Aβ suppression of Kv1.1 was partially Ca^{2+}- and PP2B-dependent, being reduced by ~50% when cells were loaded with BAPTA-AM, or exposed to the PP2B-inhibitor cyclosporine A (CsA). Patch-clamp results suggest that Aβ-suppression of Kv1.1 involves both PP2B-dephosphorylation and direct protein-protein interaction of Aβ with Kv1.1 channel subunits. Exposure of inside-out single Kv1.1 in ripped-off oocyte patches to application of purified, catalytically-active PP2B produced gradual reductions in *p*(open), followed by abrupt disappearance of Kv1.1 activity. Application of Aβ to the intracellular face of Kv1.1 channels under conditions where little enzymatic activity could occur also produced dramatic reductions in *p*(open). Additional results indicate that 2 μM of Aβ(25-35) suppressed Kv1.1 currents by ~40%. Using "tip-dip" artificial membrane methods, 1 μM Aβ(25-35) exposure eliminated Kv1.1 channel activity when applied to the intracellular face. Preliminary results indicate that "aging" the peptide at room temperature also affects suppression, with greatest suppression (~60%) seemingly occurring at 6-7 hrs for 1 μM Aβ(25-35) and 22-24 hrs for 1 μM Aβ(1-42).

Disclosures: K. Deboeuf: None. **M. Islam:** None. **N. Thelen:** None. **J. Farley:** None.

Poster

376. Potassium Channels II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 376.11/F5

Topic: B.04. Ion Channels

Title: Validation of human iPSC models of psychiatric diseases for target ID and phenotypic screening

Authors: M. NAUJOCK, V. KIZNER, S. FELK, A. SPEIDEL, S. JAEGER, G. LEPERC, K. FUNDEL-CLEMENS, T. HILDEBRANDT, C. DORNER-CIOSSEK, *B. SOMMER, F. GILLARDON
Boehringer Ingelheim Pharma GmbH & Co. KG, Biberach, Germany

Abstract: Over the past few years human induced pluripotent stem cells (iPSC) have become valuable tools for the in-vitro modeling of neuropsychiatric diseases. Starting with the neuronal differentiation of iPSC from sporadic schizophrenia patients, multiple groups reported disease associated phenotypes to proof this concept. In the present study, we differentiated 2 iPSC lines each derived from 2 healthy donors and 2 patients affected with idiopathic schizophrenia towards neuronal forebrain lineage. After around 8 weeks of total differentiation we observed spontaneous spiking and burst-firing during multi-electrode array recordings (MEA, 24- and 96-well). The human neurons also showed sensitivity to the application of neuromodulators such as bicuculline and NBQX. We further demonstrated homeostatic synaptic plasticity of iPSC-derived neurons. We then screened for disease-associated pathophysiologies using MEA and calcium imaging (FLIPR, 384-well) and observed hypoactive and hypoexcitable forebrain neurons in our schizophrenia lines. To identify underlying pathomechanisms, we performed RNA sequencing and found the potassium channel Kv4.2 subunit DPP6 being highly upregulated in neurons from schizophrenia patients. By high content imaging analysis we further observed decreased synaptic density which could be another contributor to the electrical hypoactivity. We started rescue experiments using Kv4.2 blocking compounds (AmmTx3) and lentiviral DPP6 shRNA vectors and we are minimizing the observed assay variability to enable robust phenotypic screens.

Disclosures: **M. Naujock:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co KG. **V. Kizner:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co KG. **S. Felk:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co KG. **A. Speidel:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co KG. **S. Jaeger:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co KG. **G. Leperc:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co KG. **K. Fundel-Clemens:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co KG. **T. Hildebrandt:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co KG. **C. Dorner-Ciossek:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co KG. **B. Sommer:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co KG. **F. Gillardon:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co KG.

Poster

376. Potassium Channels II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 376.12/F6

Topic: B.04. Ion Channels

Support: NIH Grant R01-DA037170

NIH Grant F30-DA039637

NIH Grant R01-DA015446

Title: Sorting Nexin 27 in midbrain dopamine neurons regulates addictive behavior to cocaine

Authors: *R. RIFKIN, J. CALLENS, J. LANDRY, G. EGERVARI, Y. HURD, P. A. SLESINGER
Neurosci., Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Exposure to addictive drugs, like cocaine, increases dopamine (DA) levels in the brain's reward circuit. G protein-gated inwardly rectifying potassium (GIRK) channels have emerged as important negative regulators of DA neuron firing. Previously, we identified Sorting Nexin 27 (SNX27) as a regulator of GIRK channels. Here, we selectively deleted SNX27 from tyrosine hydroxylase-expressing neurons ($SNX27_{TH}$ KO) and characterized the effects on $GABA_B$- and D_2-receptor activated GIRK currents. Using a retrograding, Cre-dependent virus (DIO.eYFP), we selectively recorded from ventral tegmental area (VTA) DA neurons projecting to nucleus accumbens (NAc), or substantia nigra pars compacta (SNc) DA neurons projecting to dorsal striatum (DS). In both pathways, we found that $GABA_BR$- and D_2R-GIRK currents were significantly reduced in $SNX27_{TH}$ KO mice compared to TH-$Cre^{+/-}$ controls. Further, $GABA_BR$-dependent inhibition of evoked firing was significantly decreased. Given the enhanced DA neuron excitability, we hypothesized that $SNX27_{TH}$ KO mice exhibit greater locomotor sensitization to cocaine. Mice (male/female 4-8 months) received daily IP injection of saline, followed by a subthreshold dose of 3.75 mg/kg cocaine for 5 days. Whereas 3.75 mg/kg cocaine induced little locomotor activity in TH-$Cre^{+/-}$ mice, $SNX27_{TH}$KO mice exhibited a significantly heightened response. Rescue of GIRK currents in VTA DA neurons of $SNX27_{TH}$ KO mice restored locomotor sensitization to that in control mice. We next compared self-administration of cocaine (FR1) in $SNX27_{TH}$KO mice and TH-$Cre^{+/-}$ mice. Like locomotor sensitization, $SNX27_{TH}$KO mice self-administered more cocaine (0.5 mg/kg/infusion) than TH-$Cre^{+/-}$ mice during early self-administration sessions. Similarly, $SNX27_{TH}$KO mice showed an initial higher sensitivity to self-administer cocaine at a lower dose (0.1 mg/kg/infusion) than TH-$Cre^{+/-}$ animals. Interestingly, irrespective of the dose both $SNX27_{TH}$KO and control mice eventually self-administered similar amounts cocaine at later sessions. $SNX27_{TH}$KO mice also displayed greater inactive lever-pressing activity, suggesting an underlying hyperactivity phenotype. Taken together, these experiments establish that SNX27 acts via GIRK channels in midbrain DA neurons to regulate the sensitivity to cocaine.

Disclosures: **R. Rifkin:** None. **J. Callens:** None. **J. Landry:** None. **G. Egervari:** None. **Y. Hurd:** None. **P.A. Slesinger:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.01/F7

Topic: B.07. Synaptic Transmission

Support: NIH NIBIB Grant R01EB018297

Title: Synaptic failure and functional network activity

Authors: *M. BUDAK[1], M. R. ZOCHOWSKI[2]
[1]Biophysics, Univ. of Michigan Ann Arbor, Ann Arbor, MI; [2]Physics, Univ. of Michigan, Ann Arbor, MI

Abstract: Human brain is a complex network including 10^{11} neurons connected by 10^{14} synapses. Information processing occurs in distinct, functionally specialized regions, which coordinate and integrate efficiently for the brain to function. The information transmission between neurons and brain regions occurs via synapses. Failures at these synaptic connections have detrimental effects such as loss of consciousness or neurodegenerative diseases. Our objective is understanding the effect of decreased synaptic transmission on functional connectivity of brain networks. We specifically investigate how synaptic failure affect scale free networks of interconnected integrate- and-fire excitatory neurons. Synaptic failure is introduced to the system through transmission probability, a parameter which randomly determines whether neurons transmit EPSP to other cells they're connected to or not. This parameter depends on the spiking history of the neurons, i.e. synapses are more likely to fail if the presynaptic neuron is more recently fired. The level of dependence on the synaptic history is introduced to the model with a time constant T.

We measure formation of coherent and synchronous states of activity in the network as a function of transmission probability. We demonstrate that scale-free networks with different directionalities respond to synaptic failure in different ways. However, neurons with moderate degrees are more coherent than other neurons in all scale-free network structures. We also observed that the dependence on spiking history affects synchronization and coherent state formation in different ways for different network structures. Networks with hubs of mostly outgoing connections become more coherent for an optimal range of T, then are fully disconnected with longer history dependence. These results may be applied to understand how anesthetics bring the loss of consciousness by changing brain dynamics and early diagnosis of some neurodegenerative diseases such as Alzheimer's and ALS, in which cases synaptic failure is the earliest symptom [2,3].

References

1. Bocaletti S, Latora V, Moreno Y, Chavez M, Hwang D: Complex networks: Structure and

dynamics. *Physics Reports*. 2006;424(4-5):175-308.
2. Wishart T, Parson S, Gillingwater T: Synaptic Vulnerability in Neurodegenerative Disease. *Journal of Neuropathology & Experimental Neurology.* 2006;65(8):733-739.
3. Alkire M, Hudetz A, Tononi G: Consciousness and Anesthesia. *Science.* 2008;322(5903):876-880.

Disclosures: M. Budak: None. **M.R. Zochowski:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.02/F8

Topic: B.07. Synaptic Transmission

Support: NS082650

DC013490

DC011184

DC005798

Title: Co-transmission by a basal forebrain cholinergic projection to the olfactory bulb

Authors: S. D. BURTON[1], D. T. CASE[2], J. GEDEON[2], S.-P. G. WILLIAMS[2], N. N. URBAN[2], *R. P. SEAL[2]
[1]Neurobio. and Anat., Univ. of Utah, Salt Lake City, UT; [2]Neurobio., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Basal forebrain (BF) cholinergic projections shape sensory processing in the main olfactory bulb (MOB), but the precise underlying circuitry remains unclear. Here, we report the identification of a BF cholinergic projection that innervates neurons in the MOB internal plexiform layer (IPL). Optogenetic activation of this projection elicits monosynaptic nicotinic and GABAergic currents in glomerular layer (GL)-projecting interneurons, implicating neurotransmitter co-transmission in the regulation of this inhibitory microcircuit.

Disclosures: S.D. Burton: None. **D.T. Case:** None. **J. Gedeon:** None. **S.G. Williams:** None. **N.N. Urban:** None. **R.P. Seal:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.03/F9

Topic: B.07. Synaptic Transmission

Support: ERC INTERIMPACT

Supported by the Hungarian Academy of Sciences

Title: Summation of metabotropic $GABA_B$ receptor mediated postsynaptic potentials in the supragranular layer of the neocortex

Authors: *A. OZSVÁR[1], G. KOMLÓSI[1,2], J. SZABADICS[1,3], G. OLÁH[1], G. MOLNÁR[1], G. TAMÁS[1]
[1]Univ. of Szeged, Hungarian Acad. of Sci., Res. Group For Cortical Microcircuits, Szeged, Hungary; [2]Inst. of Exptl. Medicine, Hungarian Acad. of Sci., Lab. of Thalamus Res., Budapest, Hungary; [3]Inst. of Exptl. Medicine, Hungarian Acad. of Sci., Lab. of Cell. Neuropharm., Budapest, Hungary

Abstract: Integration properties of synaptic inputs scattered over the neuronal surface are crucial in understanding input-output characteristics of neurons. Contrary to ionotropic responses however, experiments addressing the interaction of postsynaptic signals through metabotropic receptors are scarce. We investigated the summation properties of IPSPs elicited by individual neurogliaform cells, suggested to operate by volume transmission activating synaptic and extrasynaptic ionotropic and metabotropic GABA receptors. We characterized the parameters of neurogliaform cell to pyramidal cell synaptic transmission in layers 1 to 3 of the somatosensory cortex of rats with Bayesian quantal analysis and estimated the probability of vesicular release, the number of functional release sites and the size of postsynaptic potentials mediated by single vesicle release. Next, we developed a three dimensional model of all neurogliaform cells and their terminals in the upper cortical layers and calculated the number of neurogliaform cells which potentially release GABA into the same voxel of cortical volume. This model suggests that two neighbouring presynaptic neurogliaform cells represent all or a substantial portion of neurogliaform cells potentially effective in a postsynaptic voxel. We then assessed the summation of neurogliaform cell evoked postsynaptic potentials directly by recording simultaneously from three neurons and coactivated two presynaptic neurogliaform cells to evoke combined $GABA_A$ and $GABA_B$ mediated responses on pyramidal neurons. The fast, $GABA_A$ receptor mediated component of the compound responses summated slightly nonlinearly in both input combinations. In contrast, blockade of $GABA_A$ receptors revealed predominantly linear summation of the $GABA_B$ receptor mediated component of convergent IPSPs. In conclusion, metabotropic GABAergic postsynaptic interactions in the upper layers of the neocortex are

mainly linear even if all presynaptic interneurons of the same, particularly effective class in recruiting $GABA_B$ receptors, are active.

Disclosures: **A. Ozsvár:** None. **G. Komlósi:** None. **J. Szabadics:** None. **G. Oláh:** None. **G. Molnár:** None. **G. Tamás:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.04/F10

Topic: B.07. Synaptic Transmission

Title: Dendritic integration in human cortical pyramidal neurons

Authors: M. LAFOURCADE[1], L. BEAULIEU-LAROCHE[1], M. VAN DER GOES[1], E. N. ESKANDAR[2], M. P. FROSCH[3], S. S. CASH[4], *M. T. HARNETT[1]
[1]MIT, Cambridge, MA; [2]Neurosurg., Massachusetts Gen. Hosp., Boston, MA; [3]C.S. Kubik Lab. of Neuropathology, MGH, Boston, MA; [4]Dept Neurol, Mass Genl Hosp, Boston, MA

Abstract: The vast majority of data describing the functional properties of neurons and their synapses comes from animal models, mainly rodents. However, it is unclear how these results generalize to humans. This is a particularly pressing issue as animal models are increasingly being used to guide development of potential therapies for human brain disease. The degree to which these models accurately represent human neuron physiology is likely key in predicting success. We tested the hypothesis that human and rodent cortical principal neurons may exhibit differences in the biophysics of synaptic integration, as has recently been proposed. We applied whole-cell patch-clamp electrophysiology combined with two-photon imaging and glutamate uncaging to acute slices of human temporal cortex obtained from surgical resection. We first used local application of high osmolality solution to map miniature excitatory post-synaptic potentials (mEPSPs) at different dendritic locations. These values were used to guide two-photon glutamate uncaging probe branch-level input-output processing. We also measured backpropagation of action potentials (bAPs) using Ca^{2+} imaging. Despite their much larger dendritic arbors, human and rodent neurons exhibited similar input impedance, mEPSPs amplitudes, and bAPs, as well as broadly comparable dendritic spikes driven by multisite glutamate uncaging. These results suggest human neurons possess passive or active biophysical specializations counteracting the decreased excitability and enhanced cable filtering that would normally result from their larger and more elaborate morphologies.

Disclosures: **M. Lafourcade:** None. **L. Beaulieu-Laroche:** None. **M. van der Goes:** None. **E.N. Eskandar:** None. **M.P. Frosch:** None. **S.S. Cash:** None. **M.T. Harnett:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.05/F11

Topic: B.07. Synaptic Transmission

Support: Howard Hughes Medical Institute

Title: Voltage-dependence of spatial coding in hippocampal pyramidal neurons supported by persistent sodium current

Authors: *C.-L. HSU[1], X. ZHAO[1], A. D. MILSTEIN[2], N. SPRUSTON[1]
[1]Janelia Res. Campus, HHMI, Ashburn, VA; [2]Neurosurg., Stanford Univ. Sch. of Med., Stanford, CA

Abstract: The hippocampus is critically implicated in the formation and storage of episodic memories, for which spatial information is a key component. The activity of hippocampal pyramidal neurons forms a neural representation of space, which has been postulated to provide a foundation for memories. To unambiguously encode multiple memory engrams, these neurons need to adapt their coding scheme for different spatial, non-spatial and behavioral contexts. A thorough understanding of such information processing requires detailed knowledge of how the spatial selectivity of pyramidal-neuron firing arises as a consequence of circuit and neuron properties. Existing models usually assume that the spatial tuning of pyramidal neuron output is exclusively determined by the properties of the synaptic inputs they receive; however, recent evidence suggests that pyramidal neuron excitability also plays important roles. Using whole-cell patch-clamp recording in the hippocampal CA1 region of awake, freely moving rats, Lee et al. (2012; Science *337*, 849-853) found that a small modulation of somatic membrane potential can switch the functional identity of a pyramidal neuron between a silent cell (without well-defined spatially tuned synaptic responses or firing) and a place cell (with strong spatially tuned synaptic responses and firing in well-defined locations of the animal, called place fields), and these effects were rapidly reversible. Remarkably, the spatial tuning of synaptic responses observed below action potential threshold had a highly nonlinear dependence on baseline membrane potential at the soma.
To determine what kinds of cellular mechanisms support this striking voltage dependence of spatial coding, we applied patch-clamp recordings in acute brain slices (*in vitro*) and in awake, behaving mice (*in vivo*) during virtual navigation. Our experiments and computational models revealed that a voltage-gated sodium current—which strongly activates in a subthreshold voltage range and persists during depolarization over behaviorally relevant timescales—critically contributes to the voltage-dependent amplification of synaptic responses with high gain, consequently driving spatially dependent firing of CA1 pyramidal neurons upon small

depolarization. In addition, motivated by a finding that dendritic plateau potentials can induce the formation of new place fields (Nat. Neurosci. *18(8)*, 1133-1142), we found that dendritic plateau potentials were facilitated by dendritic depolarization. Therefore, we propose a model whereby depolarization influences both the readout and the formation of space-dependent activity in hippocampal pyramidal neurons.

Disclosures: C. Hsu: None. **X. Zhao:** None. **A.D. Milstein:** None. **N. Spruston:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.06/F12

Topic: B.07. Synaptic Transmission

Support: PHS Grant R01-MH-099054 (A.T.G.)

Frank and Myra Weiser Scholar Award (A.T.G.)

Title: Coexpression of AMPA and NMDA receptors reduces variability in synaptic transmission

Authors: *C. LI[1], A. T. GULLEDGE[2]
[1]Dartmouth Col., Hanover, NH; [2]Mol. and Systems Biol., Geisel Sch. of Med. At Dartmouth Col., Hanover, NH

Abstract: Glutamate is the major excitatory neurotransmitter in the vertebrate brain. AMPA and NMDA receptors (AMPARs and NMDARs) are two widely-distributed ionotropic glutamate receptor subtypes with distinct kinetics and voltage-dependencies. The efficacy of synaptic input in driving action potential generation is influenced by several factors, including distance of the synapse from the axon hillock, and the membrane potential at the time of synapse activation. Voltage attenuation in dendrites weakens distal inputs arriving at the axon, whereas membrane potential influences the driving force for synaptic current. Here, using computational simulations of simplified and realistic neuron morphologies, we show that coexpression of AMPARs and NMDARs may represent a novel intrinsic mechanism that limits variability in synaptic efficacy. When synapses contained both AMPAR and NMDAR conductances, somatic voltage responses were less dependent on dendritic location or membrane potential than were responses driven by synapses containing either conductance type alone. In ball-and-stick neurons comprising an axon, a soma, and a single passive dendrite, we simulated stochastic patterns of synaptic input along the dendrite within narrow spatial and temporal ranges to find the minimum number of co-activated synapses required to fire one action potential. Coefficients of variation in threshold synapse numbers were calculated for synapses with AMPA conductances, NMDA conductances, or both, for inputs placed at different dendritic locations or activated from different “resting”

membrane potentials. Relative to trials using AMPA or NMDA conductances alone, trials simulating both AMPA and NMDA conductances had lower coefficients of variation across resting potential and dendritic location. Similar results were found in realistic neuron morphologies: in simulated CA3 pyramidal cells and dentate granule cells, combining AMPA and NMDA conductances resulted in lower variability in the number of synapses required to generate a somatic EPSP of a given magnitude across input location and resting membrane potential. These results suggest that coexpression of NMDARs and AMPARs at excitatory synapses intrinsically limits location- and voltage-dependent variability in synaptic efficacy.

Disclosures: C. Li: None. **A.T. Gulledge:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.07/G1

Topic: B.07. Synaptic Transmission

Support: Research Unit FOR2143, funded by the German Research Foundation

German Research Foundation Grant BA1582/2-1

Schram Foundation

Title: Excitation/inhibition dendritic integration in fast spiking interneurons of the dentate gyrus

Authors: *C. ELGUETA, M. BARTOS
Freiburg Univ., Freiburg Im Breisgau, Germany

Abstract: Fast-spiking (FS) interneurons provide powerful inhibition into principal cells, enabling cortical circuits to perform complex operations. Despite their important role in network computations, we know little about how excitatory and inhibitory signals are processed in their dendritic trees. Using localized activation of glutamate and GABA receptors and single-cell computational modelling we studied integrative properties of FS interneurons in the dentate gyrus (DG) and compared it to granule cells (DGGCs). First, we observed that irrespective of their spatial distribution, summation of excitatory signals at FS interneurons was sub-linear or linear, while in contrast DG granule cells (DGGCs) showed supralinear summation. We examined how $GABA_A$ receptor-mediated inhibition interacts with excitatory inputs in both cells by analyzing the effect of localized GABA uncaging onto EPSPs evoked by glutamate microiontophoresis. We observed that in FS interneurons distal (off-path) inhibition reduced more efficiently EPSP amplitude than proximal (on-path) inhibition, in contrast in GCs proximally located inhibition was more efficient. This differential effect depended on EPSP size and on the distribution of the $GABA_A$ reversal potential, which was shown to be different for FS

interneurons and GCs as measured using perforated patch recordings. Measurements of GABA conductance combined with detailed single cell models, suggest that distal inhibition in FS interneurons is boosted by a combination of denser distal GABA conductance and a somato-dendritic gradient in membrane resistivity. Depolarizing on-path inhibition together with strong hyperpolarizing off-path inhibition in FS interneurons allowed GABAergic inhibition to control action potential generation independent of its spatial location, while in DGGCs on-path inhibition is most suited to regulate neuronal discharge. In summary we show that FS interneurons and principal cells differentially integrate excitatory and inhibitory signals at their dendrites adding another layer of complexity to neuronal processing in the DG.

Disclosures: C. Elgueta: None. **M. Bartos:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.08/G2

Topic: B.07. Synaptic Transmission

Title: Backpropagating action potentials boost dendritic inhibition

Authors: *G. TESTA-SILVA[1], M. HAUSSER[2], G. J. STUART[1]
[1]Eccles Inst. of Neurosci., The Australian Natl. Univ., Canberra, Australia; [2]UCL, London, United Kingdom

Abstract: Probing the interaction between action potentials (APs) and synaptic events is crucial to a full understanding of how neurons process their inputs. While it is commonly assumed that APs cause a complete reset of synaptic integration, we have shown this reset is variable and cell type-dependent during excitatory postsynaptic potentials (EPSPs) (Hausser et al. Science, 2001). Here, we extend these results to examine the interaction between APs and inhibitory synaptic potentials (IPSPs). Whole-cell patch-clamp recordings were made from neocortical layer 5 pyramidal neurons in slices from somatosensory cortex maintained at physiological temperatures. APs were generated antidromically, or by somatic current injection at various times relative to IPSPs simulated by somatic or dendritic current injection or evoked by extracellular stimulation. APs recorded in isolation were digitally subtracted from the combined IPSP/AP waveform to reveal their impact on IPSP amplitude and kinetics. As with EPSPs, APs timed to occur after the onset of somatic IPSPs led to a reduction in IPSP peak and integral, which was more pronounced when IPSPs were simulated by direct current injection compared to those simulated as a conductance change using dynamic clamp. In contrast, dendritic IPSPs simulated using dynamic clamp were transiently boosted by APs. Similar observations were made when IPSPs were generated by synaptic stimulation near the soma or at distal dendritic locations. Consistent with the idea that the boosting of dendritic IPSPs occurs due to an increase

in the driving force for inhibitory current flow generated by backpropagating APs (bAPs), we found that blocking AP backpropagation by local dendritic TTX application reduced boosting of dendritic IPSPs. Finally, we tested whether the boosting of dendritic inhibition by bAPs has a functional impact on the input-output relationship (f/I curve) of the neuron. Blocking bAPs with local TTX application reduced the impact of dendritic applications of GABA on the f/I curve obtained during somatic current injection, indicating that the capacity of APs to boost dendritic inhibition has functional consequences. These experiments indicate that the interaction between APs and IPSPs depends on the relative timing and location of inhibition, and that this interaction can boost the efficacy of dendritic inhibition. These effects can have a pronounced impact on the input-output relationship of pyramidal neurons.

Disclosures: G. Testa-Silva: None. **M. Hausser:** None. **G.J. Stuart:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.09/G3

Topic: B.07. Synaptic Transmission

Support: HHMI

Title: Behavioral timescale synaptic plasticity underlies place selectivity in CA1

Authors: *C. GRIENBERGER[1], K. C. BITTNER[1], A. D. MILSTEIN[2], S. ROMANI[1], J. C. MAGEE[1]
[1]HHMI/ Janelia Res. Campus, Ashburn, VA; [2]Neurosurg., Stanford Univ. Sch. of Med., Stanford, CA

Abstract: One of the most important properties of synapses is the ability to undergo activity-dependent changes in synaptic strength, or synaptic plasticity, providing a very compelling cellular model for learning and memory formation. The Hebbian rule, which represents one of the predominant models for synaptic plasticity, can be summarized as “neurons that fire together wire together”. Here we describe in hippocampal CA1 place cells a novel synaptic potentiation mechanism that is markedly different from this Hebbian rule. We found, combining experimental and computational approaches, that a complex spike, which is known to be associated with a dendritic plateau potential, was able to cause the formation of a new place field in a single trial by potentiation of synaptic inputs that arrived several seconds before and after this complex spike event. Moreover, the synaptic inputs that become potentiated were not initially coincident with any significant action potential firing or membrane potential depolarization. Thus, this potentiation mechanism, which we name “behavioral timescale synaptic plasticity”, affects inputs that were neither causal nor close in time to postsynaptic activation. The presence of such

plasticity was confirmed in slice recordings, and further experiments revealed a similar pharmacological profile for the in vitro plasticity and the in vivo place field formation. Thus, CA1 place fields are generated by a form of synaptic plasticity, which operates on a very long timescale of several seconds and is, therefore, very suitable for supporting hippocampal-dependent forms of learning.

Disclosures: C. Grienberger: None. **K.C. Bittner:** None. **A.D. Milstein:** None. **S. Romani:** None. **J.C. Magee:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.10/G4

Topic: B.07. Synaptic Transmission

Support: Grants-in- Aid for Science Research A (26250003)

Grants-in- Aid for Science Research on Innovative Areas `Mental Time` (25119004)

Title: CA1 pyramidal neurons receive spatially clustered synaptic inputs during sharp waves/ripples *Ex vivo*

Authors: *T. ISHIKAWA[1,2], Y. IKEGAYA[2]
[1]Pharmacol., Keio Univ., Tokyo, Japan; [2]Grad Sch. Pharma Sci, Univ. Tokyo, Tokyo, Japan

Abstract: Spontaneous reactivation of neurons involved in recently acquired memories during hippocampal sharp wave/ripples (SWRs) is crucial for memory consolidation. Neurons fire an action potential when the sum of synaptic inputs reaches the spike threshold. During this integrating processing, dendrites perform complex computations through their nonlinear electrical properties. The dendritic computations may depend on the spatiotemporal patterns of excitatory synaptic inputs, but it remains to be known about how synaptic inputs during SWRs are spatiotemporally orchestrated and determine neuronal output. Excitatory synaptic inputs were optically monitored *en masse* from hundreds of dendritic spines of individual pyramidal neurons in spontaneously active networks at video frame rates of 100-333 Hz while local field potentials were recorded from the CA1 pyramidal layer in cultured hippocampal slices. Spine activity significantly increased around the peak of SWs, and the SWR-locked activity was more spatially clustered on dendrites compared to baseline activity outside SWRs. The number of dendritic compartments that received clustered inputs increased during the SWR periods. These results may reify nonlinear synaptic integration in hippocampal pyramidal cells and neuronal reactivation during SWRs.

Disclosures: T. Ishikawa: None. **Y. Ikegaya:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.11/G5

Topic: B.07. Synaptic Transmission

Support: W&M Lundgrens Vetenskapsfond (#2017-1990)

Title: The effect of extracellular calcium on the E/I -ratio in pyramidal neurons in the hippocampus

Authors: *M. FORSBERG[1], H. SETH[2], A. BJOREFELDT[3], E. L. HANSE[4]
[1]Neurosci. and Physiol., [2]Univ. of Gothenburg, Gothenburg, Sweden; [3]Inst. of Neurosci. and Physiol., Gothenburg, Sweden; [4]Goteborg Univ., 40530 Goteborg, Sweden

Abstract: Ionized calcium in the extracellular space may affect evoked synaptic transmission via several opposing mechanisms. An increase in the concentration of extracellular calcium will decrease neuronal excitability, negatively affect release probability via G-protein coupled calcium sensors, and increase release probability via increased influx through voltage-gated calcium channels. The net effect of increasing the concentration of extracellular calcium is generally an increased evoked synaptic transmission, but it is unclear whether inhibitory ad excitatory synapses are equally affected.
In this study, we exposed pyramidal neurons in the CA1 region in acute slices (Wistar rats, p20-30) to two different concentrations of extracellular calcium, 1 and 2 mM. We patch-clamped the neurons and stimulated the Shaffer collaterals to evoke synaptic activity. While voltage-clamping the neurons at -70 mV we recorded EPSCs via glutamatergic AMPA receptors and while clamping at 0 mV we recorded the GABAergic IPSCs. The EPSCs and the IPSCs were decreased to the same extent when extracellular calcium was decreased from 2 to 1 mM.
One possible confounder is the presence of di-synaptic IPSCs. To investigate this we recorded the amplitude of the IPSC before and after wash in of the AMPA receptor blocker CNQX, thus getting a ratio of di-/monosynaptic inhibitory input to each cell. If extracellular calcium preferentially affects glutamatergic synapses, the neurons with less disynaptic inhibitory input would be less affected by changes in calcium. No such correlation was found. We next studied spontaneous events (sIPSC/sEPSC) and saw no changes in frequency when extracellular calcium was altered from 1 to 2 mM (sEPSCs: 0.52 ± 0.07 Hz to 0.53 ± 0.07 Hz, sIPSCs: 4.1 ± 0.51 Hz to 3.91 ± 0.32 Hz).
In conclusion, changing extracellular calcium between 1 and 2 mM does not change the E/I-ratio at synapses between CA1 and CA3 pyramidal neurons in the hippocampus.

Disclosures: M. Forsberg: None. **H. Seth:** None. **A. Bjorefeldt:** None. **E.L. Hanse:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.12/G6

Topic: B.07. Synaptic Transmission

Support: NSF IOS-1516235

NSF DMS-1547394

Title: A computational model of two-photon calcium imaging with a genetically encoded calcium indicator in spines and dendrites

Authors: B. SCHNEIDERS[1], *A. ABOUZEID[2], W. L. KATH[3]
[1]Engin. Sci. and Applied Mathematics, [3]Applied Mathematics, Neurobio., [2]Northwestern Univ., Evanston, IL

Abstract: Fluorescent calcium indicators are commonly used to image neuronal activity, as changes in calcium concentration accompany voltage-mediated events such as EPSPs, action potentials and dendritic spikes. The recently developed GCaMP family of calcium sensors has opened the door for genetically targeted, high resolution neuronal activity to be recorded in awake, behaving animals in vivo. However, interpreting the calcium signal can still be challenging.

We present a morphologically detailed computational model of the CA1 pyramidal cell, featuring calcium buffering dynamics and simulated GCaMP6 fluorescence throughout the dendrites and spines. The GCaMP construct combines green fluorescent protein (GFP) and calmodulin (CaM). We therefore model the calcium-binding kinetics of GCaMP based on those of CaM. Additionally, our model incorporates sources of calcium influx into the cytosol such as voltage-gated calcium channels, AMPA and NMDA receptors, as well as endogenous buffers and transmembrane pumps that extrude calcium from the cell.

We verify that the model reproduces a wide range of reported results from two-photon imaging experiments of CA1 pyramidal neuron dendrites and spines. In particular, the somatic, dendritic and spine compartments exhibit appropriate levels (both absolute and relative) of membrane depolarization, calcium concentration changes and indicator fluorescence in response to simulated synaptic stimulation.

Simulating GCaMP6 in dendrites provides a tool to inform our understanding of the mechanisms associated with synaptic integration by constraining the possible interpretations of calcium imaging experiments. It provides a window into the causal mechanisms that may give rise to observed calcium signals, such as sodium-mediated depolarizations, regenerative calcium-mediated events and NMDA spikes. The model also serves as a platform for generating hypotheses for further experimentation.

Disclosures: **B. Schneiders:** None. **A. Abouzeid:** None. **W.L. Kath:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.13/G7

Topic: B.07. Synaptic Transmission

Support: ANR-10-INBS-04-01

NIH MH106906

NIH 1U01NS099691

Title: Electrical structure of dendritic spines: A voltage imaging study with patterned illumination based on computer-generated holography (CGH)

Authors: J.-Y. WENG[1], D. TANESE[2], V. ZAMPINI[2], V. DE SARS[2], M. CANEPARI[3], V. EMILIANI[2], *D. ZECEVIC[1]
[1]Dept Cell & Mol. Physiol, Yale Univ. Sch. Med., New Haven, CT; [2]Neurophotonics Lab., Paris Descartes Univ., 75270 Paris, France; [3]Lab. for Interdisciplinary Physics, Univ. Grenoble Alpes, Saint Martin d'Hères, France

Abstract: Additional research is necessary to clarify the electrical structure of dendritic spines. Notwithstanding decades of theoretical and modelling studies, as well as calcium, sodium, and voltage imaging experiments and nano electrode recordings, the elementary question of the value of spine neck resistance relative to the input impedance of thin spiny dendrites is controversial. Currently, there is contradictory evidence in support and against the hypothetical electrical isolation of synapses on spine heads caused by a narrow spine neck. Such electrical isolation of synapses on spines, if present, could be responsible for a set of specific functions that would not be supported by synapses on dendrites. To obtained direct evidence, we previously used an electrochromic voltage-sensitive dye (JPW3028) as a transmembrane optical voltmeter with a linear scale capable of recording simultaneously electrical signals from individual spines and parent dendrites (Popovic, Carnevale, Rozsa, Zecevic, 2015). The results showed that the spine neck does not electrically isolate synapses on spines; electrically, these synapses behave as if they are located directly on dendrites. From these measurements, we found that the success rate (limited by light scattering and by the extent of photodynamic damage) was too low to allow studies of more complex electrical phenomena. Thus, further improvements in voltage-sensitive dye recording was necessary. Here we demonstrate a dual-color Computer Generated Holography (CGH) illumination system especially designed to perform functional recording from dendritic spines, where a first optical path is used to generate a multi-site intensity graded

patterns, allowing localization and intensities adjustment of the illumination of dendrites and spine heads, and the second path permits a simultaneous multisite uncaging. Intensity graded illumination patterns were shaped in a way to practically eliminate contamination of spine optical signals by the scattered light from the parent dendrite increasing the effective spatial resolution of functional recording. The results also suggest that the localization of the illumination exclusively on the regions of interest (spine heads and small sections on parent dendrites) can significantly reduce photodynamic damage. Finally, we showed that voltage imaging with CGH at 532 nm excitation light could be combined with patterned illumination for a simultaneous multisite glutamate uncaging.

Disclosures: **J. Weng:** None. **D. Tanese:** None. **V. Zampini:** None. **V. De Sars:** None. **M. Canepari:** None. **V. Emiliani:** None. **D. Zecevic:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.14/G8

Topic: B.07. Synaptic Transmission

Title: Electrical compartmentalization of dendritic spines prevents synaptic voltage-clamp

Authors: *L. BEAULIEU-LAROCHE, M. T. HARNETT
Brain and Cognitive Sci., MIT, Cambridge, MA

Abstract: Whole-cell voltage-clamp is routinely employed to measure synaptic conductances involved in a variety of neuronal processes, but little attention has been given to the limitations of this approach. Here, we reveal that voltage-clamp is completely ineffective for dendritic spines, the locus of most excitatory synapses in the mammalian brain. Combining simple compartmental modeling with dendritic patch-clamp recordings and two-photon optical techniques, we find that the spine neck resistance prevents voltage control of synaptic events, leading to large spine depolarization. As a consequence, voltage-clamp both substantially underestimates synaptic conductance and allows the recruitment of voltage-gated channels. These results question the utility of whole-cell voltage-clamp in studying synaptic physiology. To provide new, accurate measurements of synaptic parameters, we develop an approach to estimate AMPA conductance at single spines. Our results in cortical layer 5 pyramidal neurons indicate that single synapse AMPA conductance is much larger (IQR: 0.96-2.53 nS) than previously appreciated. Future work should consider the highly compartmentalized nature of central neurons in interrogating synaptic transmission and plasticity but also in the approaches taken to tackle those questions

Disclosures: **L. Beaulieu-Laroche:** None. **M.T. Harnett:** None.

Poster

377. Synaptic and Dendritic Integration

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 377.15/G9

Topic: B.07. Synaptic Transmission

Support: R01 NS037459

Title: Effects of focal ischemia on cortical excitation in the UCH-L1 knock-in mice

Authors: N. V. POVYSHEVA[1], F. ZHANG[2], M. E. ROSE[2], J. S. BANTON[2], S. GRAHAM[2], *G. BARRIONUEVO[1]
[1]Dept Neurosci, [2]Dept Neurol., Univ. Pittsburgh, Pittsburgh, PA

Abstract: Ischemic stroke is a devastating neurological disorder and one of the leading causes of disability in the US and all over the world. While there is a massive cell loss in the ischemic core, the neurons in the surrounding areas that were exposed to ischemia and survived can sustain chronic pathological changes both reversible and irreversible that are not well understood. We showed previously that focal ischemia results in decreased excitation in the brain as indicated by the loss of excitatory connections and slower axonal conduction velocity (Povysheva et al. 2016, SFN abstract). It was also shown that Ubiquitin C-terminal hydrolase L1 (UCH-L1) activity can protect primary neurons from hypoxic/ischemic injury induced by cyclopentenone prostaglandins (Liu et al., 2015). Here, we study the effects of focal ischemia on cortical excitation in the UCH-L1 knock-in mice to explore the neuroprotective role of UCH-L1 C152A mutation. Middle cerebral artery occlusion (MCAO), and sham surgeries were performed in UCH-L1 C152A mice as previously described (Zhang et al., 2014), and physiological properties of penumbral neurons as well as axonal conduction velocity were assessed 7 and 21 days later in brain slices. Whole-cell recordings were made from pyramidal neurons in layer 2-3 of the mouse neocortex located near to the ischemic core in the MCAO, and in similar locations in the sham mice. Spontaneous excitatory postsynaptic currents (sEPSCs) were recorded at the holding potential of -70 mV in the presence of gabazine (10 μM). Unlike in the WT, in UCH-L1 knock-in, MCAO did not result in altered amplitude and frequency of sEPSCs (ANOVA, $p>0.1$). Similarly, action potential-independent, miniature EPSCs recorded in the presence of TTX, were resistant to the MCAO in mutant mice. Axonal conduction velocity was assessed in the brain slices by placing stimulating and recording electrodes in the corpus callosum ipsilaterally and contralaterally in the MCAO affected hemisphere. Axonal conduction velocity for myelinated fibers was decreased in the MCAO as compared to sham animals (ANOVA, $p<0.001$). Importantly, this decrease was observed in 7 days MCAO, but not in 21 days. Axonal conduction velocity for unmyelinated fibers was unaffected by the MCAO (ANOVA, $p>0.1$). Thus, we

provide further evidence for the neuroprotective role of the UCH-L1 activity against the ischemic insult.

Disclosures: **N.V. Povysheva:** None. **F. Zhang:** None. **M.E. Rose:** None. **J.S. Banton:** None. **S. Graham:** None. **G. Barrionuevo:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.01/G10

Topic: B.07. Synaptic Transmission

Support: DK109204

Title: Differential regulation of calcium sensors involved in asynchronous release by $G_{i/o}$-coupled GPCRs

Authors: Z. ZURAWSKI[1], B. PAGE[2], S. T. ALFORD[3], *H. E. HAMM[4]
[1]Pharmacol., Vanderbilt Univ., Nashville, TN; [3]Dept. of Anat. and Cell Biol., [2]Univ. of Illinois at Chicago, Chicago, IL; [4]Dept. of Pharmacol., Vanderbilt Univ. Sch. of Med., Nashville, TN

Abstract: Presynaptic inhibition mediated by G protein-coupled receptors can involve a direct interaction between liberated Gβγ and the vesicle fusion machinery. In this way, Gi/o-coupled GPCRs regulate both evoked and spontaneous release through G-protein beta gamma (Gβγ) subunits. In addition, Gβγ mediates this inhibits inhibition of both synchronous and asynchronous evoked release at and downstream ofby modulating presynaptic calcium entry but also acting downstream of this calcium entry. We have previously demonstrated that Gβγ inhibits exocytosis downstream of calcium entry via competition with the fusogenic calcium sensor, synaptotagmin I (syt I) for binding sites on SNARE complexes with the calcium sensor synaptotagmin I (syt I), implicated in these forms of release. Here, we demonstrate the differential ability of Gβγ to compete with several types of calcium sensor proteins implicated in asynchronous evoked release and augmentation of release during repetitive stimulation. Gβγ is able to compete with syt I and Doc2, but not syt VII, for binding to SNAREs and inhibition ofconsequently to inhibit Ca^{2+} and calcium sensor-dependent lipid mixing and exocytosis. In contrast to syt I, which requires tens to hundreds of μM Ca^{2+} to evoke fusion, B both Doc2 and syt VII display high lipid mixing activity at μM levels of Ca^{2+} insufficient for robust syt I activity. syt Syt VII, but not Doc2, is activated by Sr^{2+} in lipid mixing assays, supporting the hypothesis that Sr^{2+}-driven asynchronous release in neurons is mediated by the synaptotagmin family of calcium sensors. Together, this these data suggests that release evoked during stimulus trains and asynchronous release evoked by different types of Ca^{2+} sensors has differential sensitivity to regulation by the Gβγ-SNARE mechanism.

Disclosures: **Z. Zurawski:** None. **B. Page:** None. **S.T. Alford:** None. **H.E. Hamm:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.02/H1

Topic: I.04. Physiological Methods

Title: 2D and 3D neuronal cultures exhibit significantly different spontaneous activity patterns

Authors: *M. HASAN[1], Y. BERDICHEVSKY[1,2]
[1]Dept. of Electrical and Computer Engin., [2]Bioengineering Program, Lehigh Univ., Bethlehem, PA

Abstract: In-vitro models of dissociated neuronal cultures are extensively used in the field of neuroscience to model disease and investigate brain functions. However, most of these models are two dimensional and cannot accommodate for the multi-layer connectivity between neurons found in the brain in-vivo. Here, we present a simple yet novel method to develop 3D neuronal culture. We used Polydimethylsiloxane to create 100 µm high micro-wells of different diameter (400-900 µm). These wells were then used to confine dense solution of cortical neurons obtained from neonatal rats. Diluted solution of same neurons was also seeded to create 2D cultures for comparison. R-GECO1 (fluorescent [Ca2+] indicator) (1) was expressed in cultures starting from first day in vitro. Optical recordings of [Ca2+] indicator (R-GECO1) showed synchronous activity across the whole culture from DIV8 for both 2D and 3D cultures. Our investigation showed significant difference between 2D and 3D neuronal culture spontaneous synchronous activity duration and interval. Effects of culture size and cell number on activity patterns were also determined. This study can potentially help the development of more effective neurological disease models and can further assist the investigation of brain functionality and neuronal circuits.

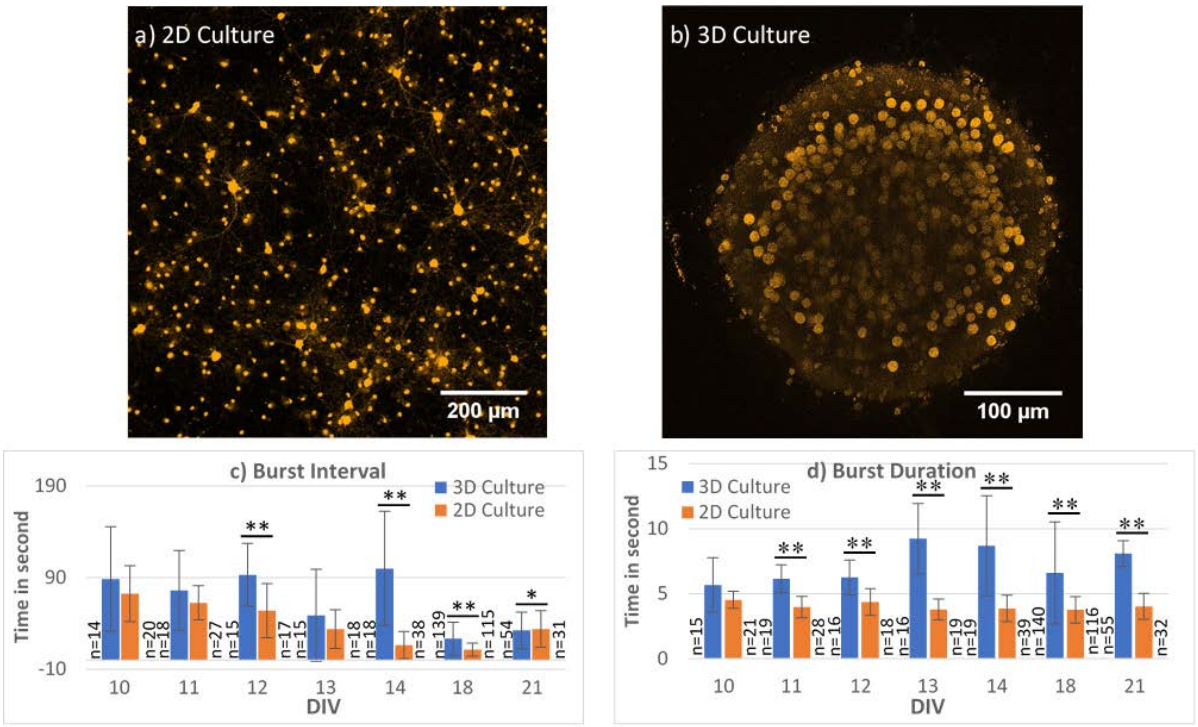


Figure: Anti-NeuN, a neuronal nucleus marker, stained **a)** 2D culture, **b)** 3D culture. Mean with standard deviation of **c)** Burst Interval, **d)** Burst Duration of 2D and 3D cultures at different days in vitro (DIV). Here, n beside each bar indicates number of intervals in (c) or durations in (d) for observed bursts on that DIV for that culture. (**= ($p<0.001$) and *=($p=0.011$) for Mann-Whitney Rank Sum Test)

(1) Yongxin Zhao et al., An Expanded Palette of Genetically Encoded Ca2+ Indicators, Year 2011, Vol. 333, pp. 1888-1891

Disclosures: M. Hasan: None. **Y. Berdichevsky:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.03/H2

Topic: B.07. Synaptic Transmission

Support: RO1MH08487

Title: Stimulus-evoked Ca^{2+} influx at a single active zone varies through a variable and small population of voltage-gated Ca^{2+} channels

Authors: *S. RODRIGUEZ[1], S. RAMACHANDRAN[3], E. C. CHURCH[2], S. T. ALFORD[4]
[2]Grad. Program in Neurosci., [1]Univ. of Illinois At Chicago, Chicago, IL; [3]Univ. Illinois, Chicago, Chicago, IL; [4]Dept. of Anat. and Cell Biol., Univ. of Illinois at Chicago, Chicago, IL

Abstract: Stimulus-evoked Ca2+ influx through voltage-gated Ca2+ channels, localized to the presynaptic release face membrane, evokes neurotransmitter release. Differing spatio-temporal Ca2+ requirements for release have been proposed in different presynaptic terminals, either gated by Ca2+ influx through many or a few open channels, with varying structural relationships to the release machinery. In most synapses, determining the variability of channel opening to stimulation is difficult because the channels and terminals are inaccessible to direct recording approaches and imaging lacks temporal and spatial resolution to determine channel activity and Ca2+ entry at active zones. We utilized the lamprey giant reticulospinal synapse to overcome these limitations. Using an acute dissociation of the lamprey spinal cord we recorded from acutely isolated reticulospinal axons, with functional presynaptic terminals devoid of apposing postsynaptic projections. Ca2+ channels were characterized at individual terminals by low-noise single channel electrophysiological recordings. We determined channel subtypes present at these terminals and estimated their probability of opening on depolarizing stimuli. The number of channels at a terminal was determined for each of the subtypes present; N-type (4-10; mean 6), P/Q-type (3-9; mean 6), R-type (4-32; mean 12) and L-type (3-17; mean 10). A small number of Ca2+ channels (up to 68, mean 33) were found to be present at a terminal. Furthermore, a very small number (3-6, mean 4) of Ca2+ channels opened in response to a stimulus, suggesting that Ca2+ influx through few open channels may gate release in these terminals. Therefore, we tested the variability of Ca2+ transients in these synapses over many active zones using high speed imaging of Ca2+ entry to the presynaptic terminal in response to an external stimulus. We found that over extended stimulation, among the active zones detected (n=12), 7 showed Ca2+ transients that did not vary beyond the variance in noise, but 5 showed distinctly variable Ca2+ transients. This variability in Ca2+ transients along with the small subset of Ca2+ channels that open in response to a stimulus suggests that there is variability in the population of Ca2+ channels that open at some active zones. This result has profound implications for the reliability and function of evoked neurotransmitter release.

Disclosures: S. Rodriguez: None. **S. Ramachandran:** None. **E.C. Church:** None. **S.T. Alford:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.04/H3

Topic: B.07. Synaptic Transmission

Title: Neuron-Specific Gene 2 (NSG2) regulates excitatory synaptic transmission via modulation of AMPAR surface expression

Authors: *P. CHANDER[1], J. P. WEICK[2]
[2]Neurosciences, [1]Univ. of New Mexico, Albuquerque, NM

Abstract: Multiple adaptor proteins have been shown to regulate the dynamic shuttling of AMPA-type glutamate receptors between intracellular compartments and the plasma membrane during synaptic plasticity. However, whether the same or novel vesicular adaptor proteins affect trafficking of AMPARs to post-synaptic membranes during synaptogenesis or under basal conditions remains unknown. Neuron specific gene 2 (NSG2) is one of the most highly expressed proteins during the period of maximal synaptogenesis in rodent and human and belongs to the Neep21/Calcyon/P19 family of endosomal proteins, but its function has remained uncharacterized. Here we show that NSG2 is found in discrete punctae restricted to somatodendritic arbors of developing neurons, and a significant proportion of NSG2 punctae co-localize with surface-expressed AMPAR subunits GluA1 and GluA2. Immunoprecipitation of overexpressed proteins in HEK293T cells, as well as from mouse brain lysates, revealed that NSG2 physically interacts with both GluA1 and GluA2. Interestingly, CRISPR-mediated knock out of NSG2 in primary hippocampal neurons selectively impaired miniature excitatory post synaptic current (mEPSC) frequency without affecting mEPSC amplitude or AMPAR surface expression. In contrast, NSG2 overexpression caused a significant increase in the amplitude of mEPSCs without significantly affecting mEPSC frequency. However, NSG2 overexpression did cause alterations in $PSD95^{+}$ expression at post-synaptic densities, suggesting a possible indirect mechanism for altering AMPAR localization and mEPSCs. Together these data show that NSG2 is an AMPAR-binding protein that regulates basal excitatory synaptic transmission during early periods of neuronal development and is required for proper synapse formation.

Disclosures: P. Chander: None. **J.P. Weick:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.05/H4

Topic: H.01. Animal Cognition and Behavior

Support: KAKENHI/16K00380,16H06532,15K0043

Title: Voltage-sesitive dye signal analysis of inhibitory components in mouse perirhinal-entorrhinal cortical slices

Authors: *Y. WAKAYAMA[1], S. KAMADA[2], Y. YAMADA[2], T. TOMINAGA[3], R. KAJIWARA[2]
[1]Electronics and Bioinformatics, [2]electronics and Bioinformatics, Meiji Univ., Kawasaki-Shi, Japan; [3]Inst. of Neuroscience, Tokushima Bunri Univ., Sanuki, KAGAWA, Japan

Abstract: The perirhinal cortex (PC) communicates with the hippocampus via the entorhinal cortex (EC). Since the PC receives diverse inputs from unimodal and polymodal association cortices, the functional linkage between the PC and the entorhinal- hippocampal network is thought to be essential for the formation and recall of memories. However, previous electrophysiological experiments in the rodents indicated that propagation of neural activity between PC and EC occurs with an extremely low probability. This phenomenon strongly suggests that inhibitory system in the PC could control the information transfer from the PC to the hippocampus via the EC. To reveal the layer distribution of inhibitory system in the PC, we analyzed the pattern of neural activities induced by electrical stimulation to superficial and deep layers of the PC respectively, on mice brain slices using voltage-sensitive dye(VSD) imaging technique. In the experiment, we perfused the 0.5 μM of gabazine, a type A gamma-aminobutyric acid (GABAA) receptor antagonist. The gabazine increased the peak value of the optical signal and prolonged the responses. To evaluate the amount of inhibitory action in the circuit, we calculated the time-integral of the optical signal (an area from baseline) at each pixel. We also calculated a pixel-wise ratio of the area before and after the perfusion. The ratio was markedly larger in the deep layer than the superficial layer. That is, the activity of the deep layer is more sensitive to the inhibition of GABAergic system than the superficial layer, thus should be under the strong control of GABAA system. Subsequently, we examined the effect of CGP-55845, a GABAB receptor antagonist. However, the CGP-55845 did not cause significant change of responses in the PC and EC. These results suggest that inhibitory mechanisms via GABAA receptors have a predominant role in the neural excitation propagation from the PC to the EC. To investigate how the excitatory response in the PC will break the wall of inhibition in the PC, we also examined the effect of the 4-aminopyridine (4-AP), which reduce the slowly inactivating potassium current (ID) of perirhinal neurons at micromolar concentrations. By the co-application of the 40 μM of 4-AP with gabazine, the stimulation to deep layers evoked large depolarized responses in EC. After washing off the gabazine, however, both deep and superficial layers stimulation in area 36 did not evoke the EC response. Our results in this study may suggest that the strong inhibitory system in deep layer of PC acting on the firing rate of perirhinal excitatory neurons may control the activation of the entorhinal-hippocampal network.

Disclosures: Y. Wakayama: None. **S. Kamada:** None. **Y. Yamada:** None. **T. Tominaga:** None. **R. Kajiwara:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.06/H5

Topic: B.07. Synaptic Transmission

Title: The glutamatergic synaptic cleft alkalinizes, rather than acidifies, during neurotransmission, a phenomenon that ameliorates depression during burst firing

Authors: *M. STAWARSKI[1], R. HERNANDEZ[2], G. T. MACLEOD[2]
[1]Dept. of Neurosci., [2]Florida Atlantic Univ., Jupiter, FL

Abstract: Recently published data give credence to the idea that the pH of the synaptic cleft is highly dynamic and that pH shifts may influence neurotransmission due to the pH sensitivity of ion channels and neurotransmitter receptors facing the cleft. However, the valence and kinetics of cleft pH transients are as of yet unknown. Using a variety of fluorescence imaging approaches, we demonstrate that cleft pH transients are spatially and temporally linked to activity-evoked postsynaptic Ca^{2+} efflux. Single action potentials evoke rapid alkalinization of the cleft of ~0.02 pH log units (~3% change in proton concentration) within ~20 ms. Although modest in amplitude, these events summate during burst firing in vivo, leading to transients that exceed 1 pH log unit. Cleft alkalinization is blocked by agents that desensitize glutamate receptors, and which also block Ca^{2+} entry to the postsynaptic compartment and its ensuing acidification. This suggests that alkaline transients in the cleft are a function of acid flux across postsynaptic membranes through plasmamembrane Ca^{2+}-ATPases. The kinetics of cleft pH transients (decaying with a time course of 60 ms) suggest that they may be relevant to fast forms of short-term synaptic plasticity. We hypothesize that activity-dependent cleft alkalinization has been incorporated into gain mechanisms that sustain neurotransmission during action potential bursts.

Disclosures: M. Stawarski: None. **R. Hernandez:** None. **G.T. Macleod:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.07/H6

Topic: B.07. Synaptic Transmission

Support: NIH Grant 5SC1MH

Title: Localization of glycinergic neurons in the mouse thalamus

Authors: *P. A. LOZANO
The Univ. of Texas At El Paso, El Paso, TX

Abstract: Glycine acts as inhibitory neurotransmitter and as a co-agonist of glutamate at the excitatory NMDA receptor in the central nervous system. Glycine Transporters (GlyT1 and 2) are responsible for the reuptake and recycling of the neurotransmitter glycine at the synaptic cleft. Recent published data and our preliminary studies suggest the presence of GlyT1 in the mouse thalamus; whether this immunoreactivity corresponds to glia cells or neurons is something that has not been clearly defined. The thalamus is considered a critical hub integrating a complex array of motor and sensory functions. The detailed anatomy of some thalamic neuronal networks, local circuits, inputs from and to the brain stem and cortex are well known. While the overall topography is well understood, the position and contribution of glycinergic networks to thalamic functions need to be elucidated. To address this question, the retrograde tracer FluroGold and two Adeno-Associated vectors expressing mCherry under synapsin or GFP under GFAP promoters, have been injected into the mouse ventral posteromedial thalamus and their distribution analyzed. Combined staining of GlyT1 with neuronal markers suggests co-localization, indicating a GlyT1 neuronal expression rather than glia cells. In addition, FluoroGold injections and labeling with GlyT1 suggest that these glycinergic neurons are interneurons rather than projection neurons. These results should shed light into the glycinergic network in the thalamus. Understanding the neural pathways containing glycine transporters will allow us to have a better knowledge about the inhibitory inputs in the control of sensory-motor integration.

Disclosures: P.A. Lozano: None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.08/H7

Topic: B.07. Synaptic Transmission

Support: NIH Grant NS064025

NIH Grant NS065920

Title: $nNOS^+$ interneurons contribute to inhibition and neurogenesis in the dentate gyrus

Authors: *A. F. MANUEL, J. C. GONZALEZ, R. J. VADEN, E. W. ADLAF, A. J. NIVER, J. I. WADICHE, L. OVERSTREET-WADICHE
Neurobio., Univ. of Alabama At Birmingham, Birmingham, AL

Abstract: The dentate gyrus (DG) is a unique brain region that exhibits very low levels of neural activity, wherein only a small percentage of principal granule cells (GCs) are active at any time, and active GCs display low spiking frequencies. The DG is also one of only two regions in the mammalian brain where adult neurogenesis occurs. GABAergic interneurons of the DG not only maintain sparse GC firing, but also provide newly generated GCs with depolarizing GABA as a trophic factor for survival and synaptic integration into the existing network. It is well established that parvalbumin (PV) expressing DG interneurons are important for DG inhibition and neurogenesis, but here we assess how a slow-spiking interneuron family called Ivy/neurogliaform cells that express neuronal nitric oxide synthase (nNOS), contribute to inhibition and neurogenesis. We crossed nNOS-CreER mice with Ai14(RCL-tdT)-D or Ai32(RCL-ChR2(H134R)/EYFP) and gave tamoxifen chow for one week after weaning. We found that all $nNOS^+$ interneurons showed slow maximal firing rates and slow action potential kinetics. $nNOS^+$ cells often had delayed spiking phenotypes, afterhyperpolarizations, and morphology characteristic of Ivy/neurogliaform cells. In contrast, PV^+ interneurons had high maximal firing rates, fast action potential kinetics, and basket cell morphology. We also crossed each ChR2-expressing mouse line with POMC-eGFP reporter mice, allowing us to record synaptic responses from both mature and newborn GCs. Optogenetic activation of $nNOS^+$ interneurons generated slow-rising and decaying $GABA_A$-mediated postsynaptic currents (PSCs) in both mature and newborn GCs that were enhanced by NO711, consistent with volume transmission from Ivy/neurogliaform cells. Single light pulses also generated $GABA_B$ PSCs in mature GCs, another hallmark of Ivy/neurogliaform-mediated transmission that differs from fast synaptic signaling by PV^+ interneurons. The combined $GABA_A$- and $GABA_B$-mediated inhibition generated by optogenetic activation of $nNOS^+$ interneurons robustly inhibited GC spiking for hundreds of milliseconds, with a slow $GABA_B$-mediated component. Furthermore, optogenetic activation of $nNOS^+$ interneurons (5Hz for four minutes, repeated every five minutes for 60 minutes), increased the number of Ki-67 labeled proliferating progenitors. Together, these data suggest that $nNOS^+$ interneurons display characteristics of Ivy/neurogliaform cells and shape both inhibition and neurogenesis in the DG.

Disclosures: A.F. Manuel: None. **J.C. Gonzalez:** None. **R.J. Vaden:** None. **E.W. Adlaf:** None. **A.J. Niver:** None. **J.I. Wadiche:** None. **L. Overstreet-Wadiche:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.09/H8

Topic: B.07. Synaptic Transmission

Support: CIHR (Canada)

Heart and Stroke Foundation (Canada)

Title: Habenula-driven protracted feed-forward inhibition mediated by 5-HT_{1A} receptors in the dorsal raphe nucleus

Authors: *M. B. LYNN[1], S. D. GEDDES[2], S. MAILLÉ[2], D. LEMELIN[2], R. BERGERON[2], S. HAJ-DAHMANE[3], J.-C. BÉÏQUE[2]
[1]Cell. and Mol. Med., Univ. of Ottawa, Ottawa, On, Canada; [2]Cell. and Mol. Med., Univ. of Ottawa, Ottawa, ON, Canada; [3]Res. Inst. on Addictions, Univ. at Buffalo, Buffalo, NY

Abstract: The habenulo-raphe pathway is believed to be involved in orchestrating behavioral responses to aversive, threatening or stressful environments. The mechanistic details of how the long-range input from the lateral habenula (LHb) to the dorsal raphe nucleus (DRN) is organized and processed is, however, largely unknown. Here, using a variety of optogenetic strategies in combination with whole-cell electrophysiology, we outlined a novel form of protracted feed-forward processing in the DRN network. We found that the LHb input to the DRN provides a powerful direct monosynaptic glutamatergic drive to 5-HT neurons that also triggers a canonical, rapid, $GABA_AR$-mediated feedforward inhibition. Intriguingly, however, train stimulation of the LHb input to the raphe also triggered a singularly distinct form of feedforward inhibition. This LHb-driven hyperpolarizing response lasted for seconds, its induction was steeply dependent on the frequency of LHb inputs and was mediated by a GIRK conductance activated by 5-HT_{1A}Rs. Optogenetic manipulations in the DRN suggest that this protracted inhibition is mediated not by a cell-autonomous autoinhibition mechanism, but rather by a feedforward inhibition enacted by 5-HT neurons organized in an unsuspected recurrent network architecture. Thus, this mechanism provides the LHb inputs with the ability to outcompete parallel inputs to the DRN. More broadly, these functional connectivity features provide an effective and perhaps generalizable neural strategy to implement a dynamic input selection mechanism in hub-like networks.

Disclosures: M.B. Lynn: None. **S.D. Geddes:** None. **S. Maillé:** None. **D. Lemelin:** None. **R. Bergeron:** None. **S. Haj-Dahmane:** None. **J. Béïque:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.10/H9

Topic: B.07. Synaptic Transmission

Support: NIH NINDS R01NS076312

F31 NS095568

Title: Elevated O-GlcNAcylation modulates inhibitory neurotransmission in hippocampal area CA1

Authors: *L. T. STEWART[1], J. C. CHATHAM[2], L. L. MCMAHON[3]
[2]Pathology, [3]Cell, Developmental, & Integrative Biol., [1]Univ. of Alabama At Birmingham, Birmingham, AL

Abstract: Studies from our lab have begun to characterize the role of protein O-GlcNAcylation on hippocampal synaptic transmission and learning and memory. This post-translational modification involves the O-linkage of β-*N*-acetylglucosamine to Ser/Thr residues on target proteins. O-GlcNAcylation is as dynamic as protein phosphorylation and is carried out by the enzyme O-GlcNAc transferase (OGT), which attaches the O-GlcNAc moiety, and O-GlcNAc (OGA) transferase, which catalyzes its removal. Together, this enzymatic set represent the final step of the Hexosamine Biosynthetic Pathway, the sole metabolic pathway for all O-GlcNAcylation in the body. Immunohistochemical staining for O-GlcNAcylated proteins in rat and human hippocampus shows pronounced immunoreactivity in all regions of hippocampus, with expression in excitatory pyramidal cells, GABAergic interneurons, and astrocytes. Using brain slice electrophysiology, we previously reported that acutely increasing O-GlcNAcylation, using either the substrate glucosamine or the OGA inhibitor Thiamet-G, induces a novel NMDAR-independent form of LTD at CA3-CA1 synapses that requires O-GlcNAc modification of GluA2 AMPAR subunits (Taylor et al., 2014). In more recent studies, we have found that increasing O-GlcNAcylation dampens ongoing epileptiform activity at these same synapses. To date, there have been no studies investigating whether GABAergic inhibition is modulated by O-GlcNAcylation, or whether this is specific for excitatory transmission. Using whole-cell voltage-clamp recordings from CA1 pyramidal cells, we investigated whether increasing O-GlcNAcylation using the OGA inhibitor Thiamet-G modulates the frequency or amplitude of spontaneous IPSCs. Preliminary experiments show a reduction in IPSC amplitude following bath application of Thiamet-G, with no change in IPSC frequency, suggesting a possible postsynaptic site of action. Further experiments will examine possible changes in evoked IPSCs onto CA1 pyramidal cells. As the balance of excitatory to inhibitory input controls the final output of CA1 pyramidal cells, E/I ratios will be measured in the presence of elevated O-GlcNAcylation. Collectively these data will further characterize the role of protein O-GlcNAcylation in modulating hippocampal function.

Disclosures: L.T. Stewart: None. **J.C. Chatham:** None. **L.L. McMahon:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.11/H10

Topic: B.07. Synaptic Transmission

Support: Jean Templeton Hugill Anesthesiology Research Fund

Title: Presynaptic actions of isovaline reduce inhibitory and excitatory postsynaptic currents in thalamic neurons

Authors: *K. A. ASSERI[1,2], B. A. MACLEOD[3], S. K. W. SCHWARZ[4], E. PUIL[4]
[1]Dept. of Anesthesiology, Pharmacol. and Therapeut., Univ. of British Columbia, Vancouver, BC, Canada; [2]Dept. of Pharmacol., King Khalid Univ., Abha, Saudi Arabia; [3]Pharmacol., Dept of APT UBC, Vancouver, BC, Canada; [4]Dept. of Anesthesiology, Pharmacol. & Therapeut., The Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Isovaline is a non-proteinogenic amino acid that inhibits thalamocortical neurons in ventrobasal nuclei. Since isovaline produces analgesia *in vivo* through activation of metabotropic glutamate group II (mGlu II) in addition to $GABA_B$ receptors, we investigated if isovaline acts on thalamic neurons in a similar fashion. We hypothesized that isovaline inhibits inhibitory and excitatory synaptic postsynaptic currents (IPSCs and EPSCs) by activating presynaptic mGlu II or $GABA_B$ receptors. Whole-cell patch clamp recordings from thalamocortical neurons of ventrobasal nuclei were performed in brain slices from Sprague-Dawley rats (P9-12). Electrical stimulation of the somatosensory input, medial lemniscus, evoked both IPSCs and EPSCs. The IPSCs and EPSCs were isolated from each other by using a holding potential that minimized outward currents (V_h = 0 mV) or inward currents (V_h = -70 mV). Kynurenic acid was applied to block inotropic glutamate currents. Internal Cs^+ was applied to eliminate postsynaptic effects of agonists of $GABA_B$ or mGlu II receptors. Bath application of the $GABA_B$ agonist, baclofen, and mGluR II agonist, LY354740, suppressed the IPSCs and EPSCs. These effects were selectively antagonized by $GABA_B$ antagonist, CGP52432, and mGlu II antagonist, LY341495, indicating the presence of $GABA_B$ and mGlu II receptors on presynaptic elements. Application of isovaline (30-1000 µM) decreased the peak amplitudes of IPSCs (EC_{50} = 284 µM; 95% CI, 76-170). Isovaline (400 µM) decreased the peak amplitudes of EPSCs by 44% (95% CI, 59-224). Isovaline did not affect the decay-time constants of IPSCs and EPSCs. Isovaline-depressions of IPSCs and EPSCs were blocked by co-application of CGP52432 or LY341495. Isovaline activation of metabotropic mGlu II and $GABA_B$ receptor subtypes on nerve terminals may affect release of glutamate or GABA. These studies demonstrate that isovaline produces inhibition of GABAergic IPSCs and glutamatergic EPSCs by actions that are mediated by metabotropic $GABA_B$ and mGlu II receptors.

Disclosures: **K.A. Asseri:** None. **B.A. Macleod:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); TherExcell Pharma. Other; Jean Templeton Hugill Anesthesiology Research Fund. **S.K.W. Schwarz:** None. **E. Puil:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); TherExcell Pharma. Other; Jean Templeton Hugill Anesthesiology Research Fund.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.12/H11

Topic: B.07. Synaptic Transmission

Support: NSF Grant IBN-0344559

NIH Grant NS58867

NIH Grant MH095948

Title: Inhibitory neurotransmission drives endocannabinoid degradation during memory consolidation

Authors: *C. DUBOIS, J. LIU
Dept. of Cell Biol. and Anat., LSUHSC, New Orleans, LA

Abstract: The endocannabinoid system represents a major regulator of brain activity by reducing synaptic transmission and neurons activity. In the cerebellar cortex, 2-AG is released on-demand from Purkinje and stellate cells and activates presynaptic CB1 receptors, which reduce the release of glutamate and GABA. MAGL is the enzyme responsible for 2-AG degradation, and thus controls the temporal profile of the endocannabinoid action. We tested the hypothesis that endocannabinoid degradation is actively regulated by neuronal activity and plays a critical role in learning. Here we show that fear conditioning increased MAGL expression in the cerebellar cortex and accelerated endocannabinoid degradation. Learning also induced a lasting increase in evoked and spontaneous GABA release from cerebellar stellate cells. Using pharmacology and optogenetics, we found that a learning-induced increase in GABA release is necessary and sufficient to enhance endocannabinoid degradation. Using DREADD technologies, we show that activation of Gq-coupled receptors in cerebellar Purkinje cells reduced GABA release in the cerebellar cortex through an endocannabinoid-dependent pathway. Pharmacogenetic activation of Purkinje cells *in vivo* after fear learning disrupted cued but not contextual learning. Therefore a learning-induced increase in GABA release accelerates 2-AG degradation in the cerebellum and promotes the consolidation of fear memory. Our finding that

fear conditioning potentiates endocannabinoid degradation by increasing MAGL expression contrasts with the widely accepted view that endocannabinoid signaling is mainly determined by “on-demand” release. This novel form of degradation-dependent plasticity remodels synaptic integration and information processing within a neuronal circuit.

Disclosures: **C. Dubois:** None. **J. Liu:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.13/H12

Topic: B.07. Synaptic Transmission

Support: NIH/NINDS R01NS053719

FONDECYT Chile #11150816

FRAXA foundation

Title: Faulty mGluR activation of the inhibition, a mechanism for homeostatic control of somatosensory cortex hyperexcitability in fragile X syndrome

Authors: *C. A. CEA-DEL RIO[1], A. F. NUNEZ-PARRA[2], S. FREEDMAN[3], D. RESTREPO[4], M. M. HUNTSMAN[3]
[1]CIBAP, Facultad de Ciencias Medicas, Univ. de Santiago de Chile, Santiago, Chile; [2]Inst. de Ciencias Biomedicas, Univ. Autonoma de Chile, Santiago, Chile; [3]Dept. of Pharmaceut. Sci., Univ. of Colorado at Denver - Anschutz Med. Campus, Aurora, CO; [4]Cell & Dev. Biology, Neurosci. Program, Univ. of Colorado Anschutz Med. Campus, Denver, CO

Abstract: Fragile-X Syndrome (FXS) patients exhibit behavioral phenotypes reflective of hyperexcitable circuitry across different brain regions. In the FXS mouse model, the disruption in the function of a the Fragile X Mental Retardation1 (*Fmr1) gene*, results in a variety of neurological consequences related to problems with the development and maintenance of synaptic function. In this study, we hypothesize that faulty activation of inhibitory interneuron subclasses by glutamate through metabotropic glutamate receptors (mGluRs) renders a major component of the inhibitory neurotransmitter system dysfunction in layer 2/3 of the somatosensory cortex. Whole-cell experiments suggests that: 1) While mGluR activation increases both sIPSC amplitude and frequency in control animals, DHPG (a mGluR agonist) application fails to increase sIPSC activity in *Fmr1* KO mice. 2) Somatostatin-positive interneurons (Sst) showed concentration-dependent mGluR activation, which triggers high frequency action potential firing in control animals. In contrast, Sst-interneurons from *Fmr1* KO mice showed a decreased action potential frequency in response to mGluR activation. 3) mGluR-

dependent plasticity mechanism of the inhibition such as slow self-inhibition (SSI) and inhibitory long term depression (LTDi) were both altered in the *Fmr1* KO mice. Thus, the overall mGluR-mediated activation of inhibitory synaptic plasticity is altered in *Fmr1* KO mice suggesting an increased inhibitory drive onto pyramidal cells. Concurrently, we also tested sensory responsiveness to stimulation of whiskers and neuronal cell activation in L2/3 of somatosensory cortex in anesthetized *Fmr1* KO mice. Tetrode recordings and single unit analysis revealed that the baseline rate of the recorded units is higher in KO mice. These are the first *in vivo* experiments showing that the cortex in *Fmr1* KO mice is hyperexcited at a basal state. Furthermore, when contralateral whiskers are stimulated with air puff, we find that subsets of cells respond differentially with either increases or decreases in firing rate in both groups. In the *Fmr1* KO, a higher percentage of units decrease their spike rate in response to whisker stimulation, suggesting the possibility of an enhanced inhibitory drive. Taken together these data suggest that neurons of the KO mice are hyperexcited and that the failure of mGluR-mediated mechanisms could compromise the normal inhibitory control of the network and further contribute as a homeostatic mechanism to the hyperactivity of the somatosensory cortex.

Disclosures: C.A. Cea-Del Rio: None. **A.F. Nunez-Parra:** None. **S. Freedman:** None. **D. Restrepo:** None. **M.M. Huntsman:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.14/I1

Topic: I.04. Physiological Methods

Support: Fondecyt 1141233

CONICYT-PCHA/Doctorado Nacional/2016-21161611

Title: *In vivo* detection of endogenous octopamine and serotonin upon conditioning-training stimuli in *Drosophila* mushroom bodies

Authors: *S. I. HIDALGO, N. FUENZALIDA-URIBE, D. F. MOLINA-MATEO, J. M. CAMPUSANO
Pontificia Univ. Católica De Chile, Santiago, Chile

Abstract: Biogenic Amines (BAs) are a group of molecules that act as neurotransmitters in the brain to modulate complex behaviors, including learning and memory formation. *Drosophila melanogaster*, an animal model with important genetic tools that shows similar mechanisms of neurotransmitter storage, release and recycling as compared to mammalian systems, has been extensively used in assays of aversive learning. In these protocols, a Pavlovian conditioning

approach is used. For instance, an electric shock is applied to flies (unconditioned stimulus, US) while they are exposed to an odorant (conditioning stimulus, CS). One of the open questions regarding this approach is what are the changes in fly brain neurotransmission during conditioning. To advance on this issue, we generated a new setup to measure the *in vivo* release of BAs in fly brain (fast scan cyclic voltammetry, FSCV) when the US and CS are applied.
A single male fly head was fixed to a recording chamber and the brain was exposed surgically. Using a carbon fiber electrode, we measured BAs release in fly brain scanning from −0.4 to 1.2 V and back (vs Ag/AgCl reference electrode; scan rate of 400 V/s at 10 Hz). As positive control for the identification and quantification of endogenous BAs in the fly brain, ATP-activated ion channels ($P2X_2$ receptors) were expressed in specific aminergic neuronal populations and activated by ATP. The electrode was placed in the Mushroom Body calycal region, identified by expression of GFP under the control of the c309-gal4 driver. Flies were exposed to a single electric shock (10 V) every 5 seconds via an electric grid. In addition, we used a vacuum pump to deliver one of two odorants; benzaldehyde or ethyl acetate.
Our results show that when ATP Is applied in flies expressing $P2X_2$ receptors in octopaminergic, tdc2-positive neurons, only octopamine is detected. When the ligand is applied in flies expressing the channel in ddc-positive neurons, the release of dopamine and serotonin is evidenced. On the other hand, release of octopamine is revealed in the Mushroom Body calyx upon application of electric shocks and mostly 5-HT is recorded upon odorant stimulation in this fly brain region. No release of BAs is detected when flies are stimulated using a nominal zero calcium solution, suggesting the amine release depends on synaptic processes.
Our results show a differential release of endogenous BAs in brains of flies exposed to stimuli relevant to olfactory learning and memory conditioning, consistent with the proposed roles for the different amines in this physiological process.

Disclosures: S.I. Hidalgo: None. **N. Fuenzalida-Uribe:** None. **D.F. Molina-Mateo:** None. **J.M. Campusano:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.15/I2

Topic: B.07. Synaptic Transmission

Support: NIH R21MH10069

NIH R01MH096274

Title: L-proline,a metabolite linked to neuropsychiatric disorders and associated with the 22q11.2 deletion syndrome, specifically disrupts high-frequency GABA-ergic transmission

Authors: *G. W. CRABTREE[1], *G. W. CRABTREE[1], *G. W. CRABTREE[1], *G. W. CRABTREE[1], ***G. W. CRABTREE**[1], J. A. GOGOS[2,3]
[1]Physiol. and Cell. Biophysics, Columbia Univ. Press, New York, NY; [2]Neurosci., Columbia Univ., New York City, NY; [3]Dept. of Physiol. and Cell. Biophysics, Columbia Univ. Med. Ctr., New York City, NY

Abstract: Accumulation of L-proline within the CNS, has been repeatedly correlated with predisposition to psychotic disorders. Proline dehydrogenase (PRODH), which degrades L-proline, resides within the 22q11.2 deletion a strong genetic risk factor for schizophrenia further suggesting L-proline maybe neuroactive and directly contribute significantly to neuropsychiatric symptomology. Previously we have shown that L-proline is GABA-mimetic. L-proline can activate both GABA-A and GABA-B receptors, however this only occurs far above CNS disease-relevant L-proline levels. At disease-relevant concentrations, L-proline GABA-mimesis appears limited to competitive blockade of glutamate decarboxylase (GAD) and causes reduced GABA production. Employing a mouse with a severely hypomorphic *Prodh* allele, we have shown with recordings from layer II/III mPFC pyramidal neurons in acute brain slices that mice with elevated CNS L-proline show isolated and specific disruptions of high-frequency GABA-ergic transmission. These deficits in GABA-ergic transmission were attributable to GAD-blockade due to elevated L-proline levels as revealed by pharmacological studies targeting GAD and GABA transaminase.
The interneuronal subtypes responsible for the observed alterations GABA-ergic transmission remain unknown although our previous results strongly suggest that parvalbumin-positive neurons (PV+) may be involved in the observed pathophysiology. Employing a PV-Cre mouse we have targeted channelrhodopsin expression to PV+ interneurons in the mPFC and performed a detailed assessment of light-evoked GABA-ergic transmission at layer II/III pyramidal neurons. Furthermore, as assessment of high-frequency synaptic transmission with channelrhodopsin can be problematic due to kinetic limitations of these channels, we employed two channelrhodopsin variants (ChETA and ChIEF) which showed marked performance differences under high-frequency stimulation. We have also initiated an exploration of GABA-ergic therapeutics that successfully rescue synaptic transmission deficits and behavioral alterations in hyperprolinemic mice. Together these findings provide evidence that deficient GABA-ergic transmission arising from PV+ interneurons may underlie the synaptic and gamma-oscillation deficits that arise due to elevated L-proline levels with wider implications for other disease-related GABA-mimetic metabolites. Further we show the promise of precisely targeted therapeutics in genetically well-defined neuropsychiatric disorders. This work was supported by NIH grants R21MH10069 and R01MH096274.

Disclosures: G.W. crabtree: None. **J.A. Gogos:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.16/I3

Topic: B.07. Synaptic Transmission

Title: Modulating effects of MDMA on spontaneous firing of the subthalamic nucleus neurons: a quantitative study

Authors: *C. MAHAPATRA[1], R. MANCHANDA[2]
[1]IIT Bombay, IIT Bombay, Powai,Mumbai, India; [2]Indian Inst. of Technol. Bombay, Mumbai, India

Abstract: MDMA has diverse psychological and physiological effects, particularly rewarding and addicting influences in both humans and animals. MDMA is considered as substrate of various neurotransmitter transporters to stimulate the efflux of neurotransmitter from vesicles into the cytosol. The subthalamic nucleus (STN) plays dominant role in several function of basal ganglia and its activity is regulated by 5-HT. This study presents a quantitative study of modulating effects of the MDMA on the firing pattern of the STN neuron via 5-HT receptors. Here the interpretation of MDMA and 5- HT are based on differential equations, where all parameters are borrowed from published data. Then, this neurotransmitter mechanism is incorporated into a published STN electrophysiological model. A brief square pulse of varied duration and magnitude is applied as an external stimulus current (I_{stim}) to trigger action potential (AP) in the whole cell model. Then MDMA concentration is varied to investigate the modulated response in AP and resting membrane potential. The single STN neuron model mimicked vigorous spontaneous action potential activity and the tonic firing pattern with regular inter-spike intervals similar to those found in experiment. The MDMA is simulated from 2 to 20 μM. We found that the mean and median firing rates in the simulation increased moderately and in a concentration- dependent manner with the application of MDMA. This finding from the simulation of STN neuron by MDMA agrees with the notion found in experimental studies, which mention that the effects of drug to its motor effects. The future extension of this preliminary study will reveal the heterogenic modulatory effects of MDMA in STN cell.

Disclosures: C. Mahapatra: None. **R. Manchanda:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.17/I4

Topic: I.06. Computation, Modeling, and Simulation

Support: NSF IGERT 0903622

College of Engineering, University of Illinois at Urbana-Champaign

ADSC, Singapore

Title: An outward potassium current (M-current) is an estimate of the neuron's input

Authors: *A. R. ASILADOR[1,2,3], E. C. JOHNSON[2,3,4], R. RATNAM[1,2,3,5,6]
[1]Neurosci. Program, [2]Beckman Inst. for Advanced Sci. and Technol., [3]Coordinated Sci. Lab., [4]Electrical & Computer Engin., [5]Hlth. Care Engin. Systems Ctr., Univ. of Illinois at Urbana-Champaign, Urbana, IL; [6]Advanced Digital Sci. Ctr., Illinois at Singapore Pte Ltd, Singapore, Singapore

Abstract: Neural spike-trains carry information about the input to the neuron in the average rate of spikes or spike-times, etc. Recently we showed that a code that maximizes the fidelity of the coding for a given long-term spike-rate (a proxy for the neuron's metabolic energy consumption), will lead to a spike-timing code. We argued that a neuron is a precise coder where each spike is timed to optimize the energy-fidelity trade-off. Coding fidelity is determined from a reconstruction of the input signal from the spike-times. A prediction of this code is that the reconstruction is ongoing with the spike generation, and manifests as a dynamic or adaptive threshold. Here we explore the biophysical mechanisms that support these ideas, and in particular postulate that the muscaranic or M-current (an outward potassium current) is the reconstruction of the input. The M-current has been implicated in spike-frequency adaptation. We tested this idea using *in vitro* current-clamp data from L5 pyramidal neurons in the rat somatosensory cortex (available from the International Neuroinformatic Coordinating Facility or INCF, Spike-time Prediction Challenge, 2009; see Gerstner and Naud, 2009). We first constructed a conductance-based (Hodgkin-Huxley type) single-neuron model with 8-10 conductances. We next determined the best values of linear and nonlinear gating and conductance parameters so that for a given injected current, the predicted time-varying membrane potential matched the experimental data. The best match to the data was determined from an optimization method proposed previously by the Abarbanel group (Meliza et al., 2014). There is good match of the sub-threshold membrane potential, spike waveform, and spike-timing with a few missed or added spikes. Analysis of currents through various channels show that the M-current is the dominant outward current that closely matches the injected current, and may

serve as a candidate decoder for reconstructing the stimulus from the ongoing spike train. Having an internal decoder allows the neuron to monitor the fidelity of coding on a moment-by-moment basis, and rapidly regulate the timing of action potentials to minimize coding error. In this process, the long-term spike rate (energy consumption) sets a bound on the coding error. These results broadly support the idea that selection pressure has most likely led to biophysical mechanisms for energy-efficient high fidelity coding.

Disclosures: A.R. Asilador: None. **E.C. Johnson:** None. **R. Ratnam:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.18/I5

Topic: I.04. Physiological Methods

Support: 1U01MH109091

Title: Light-induced activation of dopamine, glutamate and GABA release on striatal medium spiny neurons in brain slices

Authors: *E. MCKIMM, T. PASTIKA, M. B. SINGH, J. A. WHITE, S. D. ANTIC
Inst. of Systems Genomics, Univ. of Connecticut Hlth., Farmington, CT

Abstract: The nigrostriatal pathway, originating in the substantia nigra (SN) and terminating in the striatum, plays a critical role on cognitive and motor functions relevant to a variety of disorders including Parkinson’s disease, addiction, and schizophrenia. Striatal medium spiny neurons (MSNs) bear dopaminergic receptors whose activation influences behavior and cognition. A transgenic mouse with light-sensitive channels in dopaminergic neurons (DAT-ChR2) is routinely used to evaluate the physiology of dopamine (DA) neurotransmission. The present study explores the simultaneous transmission of DA, glutamate and GABA, upon blue light (475 nm) activation of DAergic axons in the striatum (brain slices) by whole-cell recordings of the postsynaptic responses in MSNs. Repetitive light activation of striatal DAergic fibers revealed a strong depression of the amino acid neurotransmitter output from these fibers, with a relative refractory period of approximately 10,000 ms. However, successive light pulses on the cell bodies of DAergic neurons in SN showed a 100-fold shorter refractory period (100 ms) for firing somatic action potentials. These differences in recovery time suggest inefficient transmission of information from DA axons to MSNs, or likely the result of severing the axons in slice preparations. A pharmacological block of DA receptors did not produce any effects on light-induced synaptic currents in MSNs. In other words, DA release appeared not to have any effect on the postsynaptic glutamate and GABA currents, although both glutamate, GABA and DA were stimulated by the same pulse of light. Further exploration of light- and synaptically-

induced DA-glutamate co-release will allow for a more thorough understanding of the complex relation between glutamate and DA on neuropsychiatric disorders influenced by the nigrostriatal pathway.

Disclosures: **E. McKimm:** None. **T. Pastika:** None. **M.B. Singh:** None. **J.A. White:** None. **S.D. Antic:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.19/I6

Topic: I.04. Physiological Methods

Support: NIH Grant EY01234

Stanford FIDL Research- 1027995-191-EAFGT

Stanford SIP Funds from OTL-1185084-100-DBIRD

Title: A new $GABA_A$ anesthetic that discriminates between tonic, fast and slow synaptic inhibition

Authors: *N. S. CAYLA[1], B. A. DAGNE[1], M. F. DAVIES[1,2], Y. WU[1], E. R. GROSS[1], M. B. MACIVER[1], E. J. BERTACCINI[1,3]
[1]Anesthesia, Stanford Sch. of Med., Stanford, CA; [2]Dva, [3]DVA, VA Hlth. Care system, Palo Alto, CA

Abstract: Background: GABA receptors are important molecular targets for anesthetics, but it remains unclear how these receptors are involved in producing unconsciousness or contributing to unwanted side effects. Our group has developed new anesthetic agents that specifically target $GABA_A$ receptors. Using an *in silico* docking-based screening program, a novel molecular core was designed to target the same binding site as etomidate and eliminate the common side effect of adrenal suppression. The top ten compounds generated by the program were first tested on tadpoles, then in rat brain slice electrophysiology experiments. Finally, *in vivo* experiments were conducted with propofol, etomidate, and the most promising compound (BB) to compare their hemodynamic effects.
Methods: For the *in vivo* studies, the compounds were pipetted into the amphibians' water and rats were administered drugs by intravenous bolus. For electrophysiology, rat brain slices were submerged in artificial cerebrospinal fluid (ACSF). In the hippocampal CA1 region, bipolar tungsten stimulating electrodes were used to evoke field potentials via Schaffer-collateral fibers. Field potentials were recorded using a microelectrode placed near the pyramidal cell body. Control recordings were acquired with brain slices in ACSF before and after compound

exposure. Additionally, we compared BB to propofol and etomidate, agents known to selectively increase $GABA_AR$-mediated inhibition, and picrotoxin, a chloride channel blocker coupled to $GABA_ARs$.
Results: When exposed to BB, tadpoles and rats quickly lost consciousness, then fully recovered. Also rats' heart rate and blood pressure appeared more stable compared to propofol's exposure. Our electrophysiology results show that BB and etomidate produced a reversible enhancement of $GABA_AR$-mediated inhibition that appeared to be more selective than propofol. All of BB's effects occurred through the specific $GABA_A$-slow receptors, like etomidate. Propofol, in contrast, clearly enhanced other forms of $GABA_AR$-mediated inhibition. The effects of both agents were fully reversed by picrotoxin.
Conclusion: BB has an anesthetic effect via $GABA_ARs$ as predicted. BB were more selective than propofol, which acts mainly on $GABA_A$-fast and -tonic receptors, instead of the $GABA_A$-slow receptors that are most sensitive to BB. This $GABA_AR$ selectivity could account for the observed decrease in undesirable side effects on hemodynamics compared to propofol. Thus, our results suggest that anesthetic action on subtypes of $GABA_ARs$ contributes to both desired and undesired effects, so more selective targeting holds great promise for developing safer anesthetics.

Disclosures: **N.S. Cayla:** A. Employment/Salary (full or part-time):; Stanford University. **B.A. Dagne:** None. **M.F. Davies:** None. **Y. Wu:** None. **E.R. Gross:** None. **M.B. MacIver:** None. **E.J. Bertaccini:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.20/I7

Topic: I.06. Computation, Modeling, and Simulation

Title: Towards a maximalist α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptor-centric biochemical model of the synapse and its application in neurodevelopmental disease research

Authors: *R. FITZPATRICK[1], U. S. BHALLA[2], M. I. STEFAN[1]
[1]Univ. of Edinburgh, Edinburgh, United Kingdom; [2]Natl. Ctr. For Biol. Sci., Bangalore, India

Abstract: α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptors are a class of ionotropic glutamate receptors primarily responsible for fast synaptic transmission in the central nervous system. Their action is critically linked to learning and memory, and are tightly regulated within the postsynaptic environment. To date, there have only been a few attempts to model AMPA receptor (AMPAR) activity at the synapse in any great detail. In most cases, these models focus on specific aspects of AMPAR function or lifecycle (e.g Gallimore, 2016, Dutta-

Roy *et al.*, 2015), and avoid simulation of the surrounding biochemical environment. This model, utilising the Complex Pathway Simulator (COPASI) modelling software, has a core of two major signalling pathway models. The first is an AMPA receptor (AMPAR) trafficking model generated by Hayer and Bhalla (2005), which also includes NMDA receptor-mediated CaMKII autophosphorylation. The second is the mGluR signalling cascade from Iyengar and Bhalla (1999). This component includes Ras/Raf activity and MAPK pathways, and connects to the other core component via PP1 and CaMKII activity. The combined model allows for mGluR activation to modulate AMPARs within a generic hippocampal pyramidal cell dendrite. This affects the rate of trafficking of receptors from the endocytic pool to the membrane to initiate stable 'high states' during pseudo-LTP situations. Additionally, the CaMKII autophosphorylation acts as a bistable switch to further retain information across days of simulation. The model thus acts to retain system state information without the maintenance of the original molecules being necessary. Our current goal is twofold. First is to build upon this to generate a comprehensive postsynaptic signalling model comprising of pre-existing and novel models. Second is to retain a focus on AMPA receptors within this model to allow for future focus on how their dynamics alter in neurodevelopmental disease states such as Fragile X Syndrome. We present here the initial undertakings of the model, showcasing the retention of each individual sub-models' published outputs, and how well the model describes other experimental work surrounding AMPAR trafficking and modulation by other pathways. These *in silico* experiments may now act as a bedrock to use this model with more explanatory analyses in mind.

Disclosures: R. Fitzpatrick: None. **U.S. Bhalla:** None. **M.I. Stefan:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.21/I8

Topic: D.01. Sensory Disorders

Support: UBACYT 2014-17, 20020130100675BA

CONICET PIP 1098

Title: Adenosine A1 receptor agonist induces gene expression changes in light induced retinal degeneration

Authors: M. SOLIÑO[1], I. M. LARRAYOZ[2], E. M. LÓPEZ[1], M. REY-FUNES[1], A. MARTÍNEZ[2], E. GIRARDI[1], *J. J. LOPEZ-COSTA[1]
[1]IBCN, Univ. De Buenos Aires-Conicet, Buenos Aires, Argentina; [2]Angiogenesis Study Group, Ctr. for Biomed. Res. of La Rioja (CIBIR), Logroño, Spain

Abstract: A1 receptor (A1R) has been described in the retina and has been linked to the control of the characteristics of the subretinal space. A1R plays a role in inflammation, neuronal and trophic factor activity. Recently, in our lab, the modulation of A1R emerged as an effective neuroprotective strategy of the retina in the model of light induced retinal degeneration (LIRD). In order to further characterize the neuroprotective effects underlying A1R agonist treatment, mRNA changes induced by N6-cyclopentyladenosine (CPA), an A1R agonist, were studied in the retina using a LIRD model. Sprague Dawley rats received a single intravitreal injection of either CPA or vehicle, and then were subjected to continuous illumination (CI) (12000 lux) during 1 day. After this, the retinas were extracted and processed by RT-PCR. We selected as genes of interest adenosine receptors (A1, A2a, A2b and A3), proangiogenic factors (adrenomedullin and VEGF), an antiangiogenic factor (PEDF), inflammation mediators (IL-1b, iNOS, nNOS, and TNF) and a glial reactivity marker (GFAP). Data were statistically analysed using t-test on the Graphpad Prism software. RT-PCR results showed a significant decrease of A1, A2a, and A2b receptor mRNA in the retina of CPA treated eyes ($p<0.05$ in every case) while no changes in A3 mRNA were observed. A strong trend ($p=0.0515$) towards a decrease in adrenomedullin mRNA was observed), while VEGF expression did not change. Surprisingly, PEDF mRNA also decreased significantly ($p<0.05$) in CPA treated eyes. IL-1b, iNOS, and TNFα also showed a tendency to lower levels of expression in the retinas of CPA treated eyes although they were not statistically significant. nNOS mRNA did not change. GFAP mRNA was also significantly reduced by CPA ($p<0.05$). Our results show that CPA, an A1 agonist, is able to prevent the retinal damage induced by excessive light exposure. We believe that this is, at least, partially accomplished by a shift in the regulation of gene expression at the transcriptional level of diverse genes implicated in adenosine mediated transmission, angiogenesis, inflammation, and glial reactivity. Obviously, other genes such as those encoding programmed cell death or survival and/or proliferation signals may be also affected. Further research is needed to test to the full extent the potential therapeutic effects of A1R in retinal degenerations, a set of still untreatable maladies.

Disclosures: **M. Soliño:** None. **I.M. Larrayoz:** None. **E.M. López:** None. **M. Rey-Funes:** None. **A. Martínez:** None. **E. Girardi:** None. **J.J. Lopez-Costa:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.22/I9

Topic: I.04. Physiological Methods

Support: NIH Grant MH094839

NIH Grant NS084473

McKnight Foundation

Title: A dynamic clamp on every rig

Authors: *N. S. DESAI, R. GRAY, D. JOHNSTON
Ctr. for Learning and Memory, Univ. of Texas at Austin, Austin, TX

Abstract: The dynamic clamp should be a standard part of every cellular electrophysiologist's toolbox. That it is not, even 25 years after its introduction, comes down to three issues: money, the disruption adding dynamic clamp to an existing patch clamp rig entails, and the technical prowess required of experimenters. These have been valid and limiting issues in the past, but no longer. Technological advances associated with the so-called "maker movement" render them moot. We demonstrate this by implementing a fast (>20 kHz) dynamic clamp system using an inexpensive microcontroller (Teensy 3.6). Assembling the system requires no prior electronics experience and modifying it - for example, to add Hodgkin-Huxley-style conductances - requires no prior programming experience. The system works together with existing patch clamp data acquisition systems (for Macintosh, Windows, and Linux); it does not attempt to supplant them. Moreover, the process of assembling, modifying, and using the system constitutes a useful pedagogical exercise for students and researchers with no background but an interest in electronics and programming. We demonstrate the system's utility by implementing conductances as fast as a transient sodium conductance and as complex as the Ornstein-Uhlenbeck conductances of the "point conductance" model of synaptic background activity.

Disclosures: N.S. Desai: None. **R. Gray:** None. **D. Johnston:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.23/I10

Topic: I.04. Physiological Methods

Support: DOE LDRD 16-ERD-035

Title: Fast-scan cyclic voltammetry to differentiate structurally similar monoamines

Authors: *A. M. BELLE, B. J. BELMONT, A. M. YORITA, C. SUPIN
Materials Engin. Dept., Lawrence Livermore Natl. Lab., Livermore, CA

Abstract: Fast-scan cyclic voltammetry is an established technique that allows for selective, quantitative electrochemical detection of many key neurotransmitters including but not limited to dopamine (DA) and norepinephrine (NE). But, even with the application of specialized voltammetric waveforms and the addition of selective membranes to the electrode surface the

technique cannot differentiate structurally similar molecules. For example, despite the very distinct functions they serve in the body, the difference structurally between DA and NE is only a single hydroxyl group. Here we present a method that allows fast-scan cyclic voltammetry to determine and differentiate a pure or mixed monoamine signal. This differentiation occurs through the application of different voltammetric waveforms to four distinct electrodes in close proximity to one another followed by interpretation of signal ratios from the electrode array. The use of additional electrodes and waveforms allows us to separate otherwise similar signals through use of slight differences in kinetics and adsorption of two very similar molecules. In vitro results show that there are differences in the sensitivity of different voltammetric waveforms to NE and DA. Through optimization of the waveforms used with this technique the signal ratios from all waveforms create a distinct signature for both NE and DA. Use of these distinct ratios allows in vitro determination of the purity of a DA or NE signal. Finally, use of singular value decomposition analysis combined with this multi-waveform, multi-electrode technique shows promise for its use differentiating DA and NE in vivo as well.

Disclosures: **A.M. Belle:** None. **B.J. Belmont:** None. **A.M. Yorita:** None. **C. Supin:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.24/J1

Topic: I.04. Physiological Methods

Support: Howard Hughes Medical Institute

Title: Reconstructing circuit development in the zebrafish embryonic spinal cord at the single-cell level

Authors: *Y. WAN[1], Z. WEI[2], S. DRUCKMANN[3], P. KELLER[4]
[1]HHMI Janelia Res. Campus, Ashburn, VA; [2]Janelia Res. Campus, Ashburn, VA; [3]Howard Hughes Med. Inst. Janelia Farm Res. Campus, Ashburn, VA; [4]HHMI Janleia Res. Campus, Ashburn, VA

Abstract: Spontaneous, patterned neuronal activity has been closely linked to developmental mechanisms shaping the early nervous system and is suggested to play a key role in the fine-tuning of developing circuits. However, little is known about how patterned activity emerges de novo and what factors control this maturation process. The lack of functional recordings at the whole-circuit level with single-neuron resolution and throughout the period over which patterned activity emerges has been a major limitation to understanding the roles of individual neurons in circuit formation. Here, we use Simultaneous Multi-view (SiMView) Light-sheet Microscopy to image the emergence of patterned activity in the developing spinal cord of embryonic zebrafish

and mapped calcium activity in all post-mitotic neurons for a large fraction of the developing spinal cord at a temporal resolution of 4 Hz. These data show that spinal cord neurons undergo a rapid transition from sporadic single-neuron activity to ipsi-laterally correlated and contra-laterally anti-correlated activity between 18 and 22 hours post fertilization. We developed a computational model to reconstruct the maturation process of this spinal cord circuit from our image data at the single-neuron level, characterized dynamic changes in functional connectivity as a function of time, and mapped the process onto a digital anatomical atlas. We found that early activity first emerges in neurons with conserved segmental distribution, and neighboring communities subsequently merge by synchronization to form a patterned network. Finally, we identified the cell types of active neurons using genetic and histological markers and found that different types of neurons play distinct roles in circuit maturation. Motor neurons appear to serve as pioneers in the emergence of local functional communities, whereas dorsal commissural neurons may play a key role in establishing and maintaining the phase-locked state between left and right hemi-segments of the spinal cord. The function of glycinergic commissural neurons was further confirmed by ablation and characterization of genetically manipulated fish with abnormal glycinergic transmission. By combining this characterization of the functional maturation process with developmental imaging we furthermore established a method for systematically studying the role of cell lineages and embryonic morphogenesis in the context of circuit formation.

Disclosures: **Y. Wan:** None. **Z. Wei:** None. **S. Druckmann:** None. **P. Keller:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.25/J2

Topic: D.01. Sensory Disorders

Support: World Class Institute (WCI) Program of the National Research Foundation of Korea (NRF) funded by the Ministry of Education, Science and Technology of Korea (MEST) (NRF Grant Number: WCI 2009-003)

NIH grants DC005259 and NS099691.

Title: Imaging membrane potential of the endoplasmic reticulum with a genetically-encoded voltage indicator

Authors: *M. SEPEHRI RAD[1], L. B. COHEN[1,2], B. J. BAKER[1]
[1]Korea Inst. of Sci. and Technol., Seoul, Korea, Republic of; [2]Yale Univ. Dept. of Cell. and Mol. Physiol., New haven, CT

Abstract: In eukaryotic cells, the endoplasmic reticulum (ER) is the largest continuous membrane-enclosed network which surrounds a single lumen. Using a newly designed genetically encoded voltage indicator (GEVI), we applied the patch clamp technique to HEK293 cells and found that there is an electrical interaction between plasma membrane and ER membrane. We have optically monitored the voltage changes in both of these membranes simultaneously. The optical signal of the GEVI in the plasma membrane is consistent from trial to trial. However, the ER signal decreases in size with repeated trials while the plasma membrane resistance remains constant. This dynamic behavior of the internal signal suggests that voltage may stress the ER causing it to remodel and change its resistance. Our findings further suggest that the ER may transfer electrical signals from the plasma membrane to the nuclear envelope.

Disclosures: **M. Sepehri Rad:** None. **L.B. Cohen:** None. **B.J. Baker:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.26/J3

Topic: I.06. Computation, Modeling, and Simulation

Support: Hopstem biotechnology

Title: Human iPS cells-derived neurons as platforms for drug discovery and screening

Authors: *J. FAN, T. ZOU, Q. LIU, Y. MEI
Hopstem Biotech. Ltd. Co, Zhejiang, China

Abstract: There has been needs for better models of neurological and neurodegenerative disorders besides rodents and other animal models. Human iPS cell (hiPSc) and hiPSc-derived neuronal models have emerged as novel cell models and drug screening platforms for these disorders. We have established multiple human iPS-derived neural and organoid models for autism, schizophrenia, epilepsy and Alzheimer's disease, etc.Our neural networks fully represent the excitatory and inhibitory balance of human brain and contain PV, nNOS and other interneurons that are critical for these disorders.Comparing to control hiPSc-derived neural networks, these patient hiPSc-derived neural networks show siginificant deficits in mutltiple assays and readouts. Our hiPSc neural models will greatly promote drug and biomarker discovery for these neurological disorders.

Disclosures: **J. Fan:** A. Employment/Salary (full or part-time):; Hopstem Biotechnology Ltd. Co. **T. Zou:** A. Employment/Salary (full or part-time):; Hopstem Biotechnology Ltd. Co. **Q.**

Liu: A. Employment/Salary (full or part-time):; Hopstem Biotechnology Ltd. Co. **Y. Mei:** A. Employment/Salary (full or part-time):; Hopstem Biotechnology Ltd. Co.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.27/J4

Topic: I.06. Computation, Modeling, and Simulation

Support: Italian Ministry of Health RC1604IT46

Italian Ministry of Health RC1604IS45

Italian Ministry of Health RC1601MI11

Revert Onlus

Title: Establishment of stable, expandable and safe iPSCs derived-human neural stem cell lines suitable for cell therapies and disease modeling

Authors: J. ROSATI[1], D. FERRARI[2], F. ALTIERI[1], S. TARDIVO[1], C. RICCIOLINI[3], C. FUSILLI[1], C. ZALFA[2], D. C. PROFICO[1], L. BERNARDINI[1], E. VALENTE[4], E. BINDA[1], M. COPETTI[1], G. LAMORTE[1], T. MAZZA[1], M. GELATI[1,3], A. SIMEONE[5], *A. L. VESCOVI[2,1,3]
[1]I.R.C.C.S Casa Sollievo della Sofferenza, San Giovanni Rotondo (FG), Italy; [2]Dept. of Biotech. and Biosci., Univ. of Milan Bicocca, Milano, Italy; [3]Lab. Cellule Staminali, Cell Factory and Biobanca, Terni, Italy; [4]Hosp. Santa Lucia, Rome, Italy; [5]Consiglio Nazionale delle Ricerche, Inst. of Genet. and Biophysics, Naples, Italy

Abstract: Establishing specific cell lineages from human induced pluripotent stem cells (ihPSCs) is vital for novel cell therapy approaches in regenerative medicine, particularly neurodegenerative disorders. Human neural precursors have been induced from ihPSCs. The establishment of human neural stem cells from ihPSCs (ihNSCs), with characteristics that are vital to warrant cGMP certification and clinical usage and that match those of allogeneic fetal hNSCs used in phase I/II clinical trials, has yet to be accomplished. By a virus-free technique, we established ihNSCs whose characteristics were compared and confirmed to overlap with those of the clinical grade hNSCs, successfully used in a phase I clinical trial on ALS patients. iPSCs lines were generated by the nucleofection of adult fibroblasts with episomal vectors that do not integrate into the host genome, and subsequently neuralized by using a chemically defined, serum free medium that permitted the exclusive expansion of the hNSCs. The 5 ihNSCs lines that fulfill all the functional requirements to be considered *bona fide* stem cells according to the Neurosphere Assay (long-term proliferation, self-renewal and multipotency). These lines steadily maintain their functional properties after extensive *in vitro* manipulation (up to 25

passages), critically depend on EGF and FGF2 for extensive self-renewal and spontaneously differentiate into astrocytes, oligodendrocytes and neurons upon their removal ex-vivo. Whole transcriptome sequencing of ihNSCs, followed by stringent bionformatic scrutiny, confirmed the neural and stem identity of these cells. Karyotype stability over passages and long term transplantation studies (6 months) into the brain of immune-deficient animal, have further confirmed the safety and non-tumorigenicity of ihNSCs. To validate the use these cells for therapeutic purposes we have molecularly and functionally compared the ihNSCs with clinical grade, fetal brain-derived hNSCs and showed that the proliferation rate, the differentiation abilities and safe integration into the CNS are coinciding between the hNSCs derived from the two sources.
This technique, which will be promptly translated into a cGMP compliant setting, paves the way to the establishment of clinical-grade certified, ihNSC lines for standardized clinical trials.

Disclosures: **J. Rosati:** None. **D. Ferrari:** None. **F. Altieri:** None. **S. Tardivo:** None. **C. Ricciolini:** None. **C. Fusilli:** None. **C. Zalfa:** None. **D.C. Profico:** None. **L. Bernardini:** None. **E. Valente:** None. **E. Binda:** None. **M. Copetti:** None. **G. Lamorte:** None. **T. Mazza:** None. **M. Gelati:** None. **A. Simeone:** None. **A.L. Vescovi:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.28/J5

Topic: B.07. Synaptic Transmission

Support: NSERC RGPIN386664

Title: Differential input resistance underlies asymmetrical electrical coupling between identified cardiorespiratory neurons

Authors: G. Z. ZHU[1], A. AZIZ[1], Y. GU[1], *N. S. MAGOSKI[2]
[1]Biomed. and Mol. Sci., [2]Queen's Univ., Kingston, ON, Canada

Abstract: Electrical transmission ensures firing synchrony and recruitment within neuronal circuits. Coupling is often symmetrical, where the extent to which a voltage change in one neuron alters the voltage of a second neuron is the same in both directions. However, coupling can also be asymmetrical, where the postsynaptic voltage change is different depending on the direction. Possible mechanisms for asymmetry include differential junctional conductance or differential input resistance. The present study examines asymmetrical coupling between two neurons from the freshwater pond snail, *Lymnaea stagnalis*. The neurons in question, identified as Visceral Dorsal 1 (VD1) and Right Parietal Dorsal 2 (RPD2), are tonically active, innervate the heart, mantle, and pneumostome, and synthesize both a met-enkephalin-like peptide as well

as so-called VD1/RPD2 peptides. Dual sharp-electrode current-clamp recordings from isolated brains found that the VD1 to RPD2 coupling coefficient was greater than that of RPD2 to VD1. Prior work showed the junctional conductance between VD1 and RPD2 to be equal in both directions; thus, we examined input resistance and observed that VD1 was more resistive than RPD2. If the neurons were hyperpolarized, both the input resistance of RPD2 and the RPD2 to VD1 coupling coefficient increased, while the VD1 resistance or the VD1 to RPD2 coupling remained stable. To employ a more physiological stimulus, dopamine was pressure-applied to the soma of either VD1 or RPD2. This resulted in transient inhibition of a given neuron, which also transferred to the coupled partner asymmetrically; similar to the coupling coefficients, this transfer of inhibition was greater from VD1 to RPD2 than vice versa. These data suggest that the VD1-RPD2 asymmetrical coupling is due to differential input resistance, where current, either from the recording electrode or the dopamine response, arising in the higher-resistance VD1 flows more readily through the junction to RPD2. Hyperpolarization renders the synapse symmetrical by causing a selective increase in RPD2 resistance and subsequently promoting current flow to VD1. The role of VD1 and RPD2 is to provide a constant stimulatory drive to the heart and various respiratory organs, with VD1 serving as the master oscillator and RPD2 as the follower. Asymmetrical coupling may guarantee that at resting membrane potential, or at least in a manner analagous to coupling coefficient, interspike membrane potential, RPD2 always follows the output of VD1.

Disclosures: **G.Z. Zhu:** None. **A. Aziz:** None. **Y. Gu:** None. **N.S. Magoski:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.29/J6

Topic: B.07. Synaptic Transmission

Support: FWF Austrian Science Fund

Title: A synthetic neurotransmission system to selectively modulate synaptic strength with intrinsic encoding and connectivity

Authors: *C. MCKENZIE, H. JANOVJAK
IST Austria, Klosterneuburg, Austria

Abstract: A major challenge in neuroscience research is to dissect the circuits that orchestrate behavior in health and disease. Proteins from a wide range of non-mammalian species, such as microbial opsins, have been successfully transplanted to specific neuronal targets to override their natural communication patterns. The goal of our work is to manipulate synaptic communication in a manner that closely incorporates the functional intricacies of synapses by

preserving temporal encoding (i.e. the firing pattern of the presynaptic neuron) and connectivity (i.e. target specific synapses rather than specific neurons). Our strategy to achieve this goal builds on the use of non-mammalian transplants to create a synthetic synapse. The mode of modulation comes from pre-synaptic uptake of a synthetic neurotransmitter (SN) into synaptic vesicles by means of a genetically targeted transporter selective for SN. Upon natural vesicular release, exposure of the SN to the synaptic cleft will modify the post-synaptic potential through an orthogonal ligand gated ion channel. To achieve this we have functionally characterized, in human embryonic kidney (HEK) 293 cells, primary dissociated neurons, and co-cultures, a mixed cationic methionine (met) gated ion channel from Arabidopsis thaliana and multiple prokaryotic uptake systems that are substrate specific for met. Synthetic synapses will provide a unique opportunity to manipulate synaptic communication while maintaining the electrophysiological integrity of the pre-synaptic cell. In this way information may be preserved that was generated in upstream circuits and that could be essential for concerted function and information processing.

Disclosures: C. McKenzie: None. **H. Janovjak:** None.

Poster

378. Neuronal Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 378.30/J7

Topic: B.09. Physiological Properties of Neurons

Title: Reduction of the crayfish caudal photoreceptor firing rate during anesthesia

Authors: *J. R. DEARWORTH, Jr., S. C. NESBIT
Lafayette Col., Easton, PA

Abstract: Ketamine and pentobarbital are two anesthetics that have been shown to inhibit activity of crayfish cells, whereas xylazine is an anesthetic that had not previously been tested in the animal. Xylazine was predicted to reduce action potential firing rate because the drug exhibits its anesthetic effects by blocking calcium ion flow into the neuron. One type of neuron that was used to test the effects of xylazine anesthesia in the crayfish was the caudal photoreceptor. Two of these cells are located in the crayfish sixth abdominal ganglion and they respond to light presentation by increasing firing rate and are thought to be involved in circadian rhythmicity. The caudal photoreceptor light response served as an assay to compare the effects of the aforementioned drugs on the non-image-forming visual system. Doses of drugs were first determined by their ability to block cheliped closure and eyestalk withdrawal reflexes. ED_{100} of xylazine, ketamine, and pentobarbital were 128 mg/kg, 125 mg/kg, and 160 mg/kg, respectively. *In vitro* extracellular electrical recording done in 60 crayfish showed administration of anesthetics at ED_{100} reduced the caudal photoreceptor firing rate during light presentation. In

addition, latency of light response was increased by ketamine administration, but spontaneous activity was not affected. These results suggest that anesthetic administration in the crayfish can interfere with functions of the caudal photoreceptors.

Disclosures: J.R. Dearworth: None. **S.C. Nesbit:** None.

Poster

379. Homeostatic Synaptic Plasticity: Cellular and Model Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 379.01/J8

Topic: B.08. Synaptic Plasticity

Support: Wellcome Trust Sir Henry Wellcome Postdoctoral Fellowship (EG)

Title: Integrated plasticity in dopaminergic neurons

Authors: *E. GALLIANO[1,2], V. N. MURTHY[3,4], M. S. GRUBB[5]
[1]King's Col. London, London, United Kingdom; [2]Mol. and Cell. Biol., [4]Ctr. for Brain Sci., [3]Harvard Univ., Cambridge, MA; [5]MRC Ctr. Dev. Neurobiol., London, United Kingdom

Abstract: One of the key questions in modern neurobiology is how the environment influences the brain. How does the brain respond to alterations in experience to plastically modify itself at the cellular level? Since the first demonstration of functional synaptic plasticity, two additional broad classes of neuronal plasticity - intrinsic and structural - have been characterized. Up to now these processes have largely been studied in isolation. Neurons,however,do not singularly adopt plasticity mechanisms, and it has become of primary importance to understand how they combine within individual cells and how they impact information processing in neuronal networks. Here we describe an integrated synthesis of sensory-induced neuronal plasticity - structural, intrinsic and synaptic - in a well-defined cell population: dopaminergic (DA) neurons in the olfactory bulb (OB). Bulbar DA neurons are extremely plastic, and act at a crucial point in the olfactory pathway to inhibitinformation transmission from nose to cortex. Using a reliable, physiologically-relevant and reversible means of altering sensory experience, we compared activity-dependent plasticity in OB DA neurons in juvenile mice subjected to 24h unilateral naris occlusion. We combined *ex vivo* immunohistochemistry, *in vitro* patch-clamp recordings and *in vivo* functional imaging to uncover activity-dependent structural modifications of the axon initial segment that are tightly linked with intrinsic firing properties, and are associated with striking decreases in odour-evoked activity. These integrated plastic changes at the cellular level have the potential to underlie network-level compensatory adaptation to alterations in sensory input, allowing maintained levels of sensory performance in an ever-changing world.

Disclosures: E. Galliano: None. **V.N. Murthy:** None. **M.S. Grubb:** None.

Poster

379. Homeostatic Synaptic Plasticity: Cellular and Model Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 379.02/J9

Topic: B.09. Physiological Properties of Neurons

Title: Cell-autonomous homeostatic mechanisms activated by activity deprivation

Authors: *V. VALAKH, S. NELSON
Biol., Brandeis, Waltham, MA

Abstract: Homeostatic regulation of neuronal function is an important form of plasticity that allows for stability of the brain circuit throughout development and in response to activity perturbation. The importance of these changes is illustrated by the network response to chronic activity deprivation. *In vivo*, as well as in organotypic slice culture model, prolonged activity deprivation via tetrodotoxin (TTX) causes the network to develop frequent, highly synchronous bouts of activity. This network response to global activity withdrawal is accompanied by profound changes in cortical excitatory synaptic input and inhibitory synaptic input, as well as intrinsic excitability of both inhibitory and excitatory neurons. While some of these changes, such as excitatory synaptic input, are known to be driven cell-autonomously in excitatory cells, the time scale and whether other synaptic and intrinsic changes in excitatory and inhibitory cell types are similarly regulated is much less clear. The focus of this research is to understand the mechanisms that drive each of these changes and whether cell-autonomous silencing of individual neurons is sufficient to drive this form of plasticity. We have employed optically-activated anion channels (Govorunova et al., 2015) to reversibly silence individual cells in an otherwise non-perturbed network. Since the organotypic slice culture network develops spontaneous bouts of activity, termed Up states, it is important for the inhibition to be strong enough to prevent the cell from firing even during the strong excitatory input the cell receives during one of those Up states. Indeed, during light stimulation, the cells expressing the anion channel rhodopsin do not fire action potentials in response to a series of depolarizing steps or during an Up state, while the rest of the network remains intact. Upon 3 days of continuous light activation, silenced pyramidal neurons become more excitable demonstrating that changes in intrinsic excitability are driven by cell-autonomous mechanisms and 3 days of silencing is sufficient. Further experiments will determine whether inhibitory synaptic input is also regulated cell-autonomously and whether the same type of regulation exists in other cell types.

Disclosures: V. Valakh: None. **S. Nelson:** None.

Poster

379. Homeostatic Synaptic Plasticity: Cellular and Model Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 379.03/J10

Topic: B.08. Synaptic Plasticity

Support: CIHR Grant MOP115061

Title: Metaplasticity at CA1 synapses by homeostatic control of presynaptic release dynamics

Authors: *J.-C. BEIQUE[1], C. SOARES[2], K. F. LEE[3]
[1]Cell. and Mol. Med., [3]Cell. & Mol. Medicine, RGN3501N, [2]Univ. of Ottawa, Ottawa, ON, Canada

Abstract: Hebbian and homeostatic forms of synaptic plasticity operate on different time scales to regulate synaptic strength. The degree of mechanistic overlap between these plasticity processes, and their mutual influence, are still incompletely understood. Here, we found that the homeostatic synaptic strengthening that develops in response to prolonged network inactivity was accompanied by a compromised ability of CA1 synapses to exhibit LTP. This effect could not be accounted for by an obvious deficit in the postsynaptic capacity for LTP expression, since neither the fraction of silent synapses nor the ability to induce LTP by two-photon glutamate uncaging were reduced by the homeostatic process. Rather, optical quantal analysis revealed that homeostatically strengthened synapses displayed a markedly reduced capacity to maintain glutamate release fidelity during repetitive stimulation, ultimately impeding the induction, and thus expression, of LTP. By regulating short-term dynamics of glutamate release, the homeostatic process thus influences key features of dynamical network function and exhibits features of metaplasticity.

Disclosures: J. Beique: None. **C. Soares:** None. **K.F. Lee:** None.

Poster

379. Homeostatic Synaptic Plasticity: Cellular and Model Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 379.04/J11

Topic: B.08. Synaptic Plasticity

Support: NIH, NINDS Grant R21NS084358

NIH, NINDS Grant R01NS065992

Title: GABAergic and AMPAergic synaptic scaling appear to have different trigger mechanisms in mouse cortical neurons

Authors: *C. E. GONZALEZ-ISLAS[1,3], P. BUELOW[4], P. A. WENNER[2]
[2]Physiol., [1]Emory Univ. Sch. of Med., Atlanta, GA; [3]Ciencias Biológicas, Univ. Autonoma de Tlaxcala, Tlaxcala, Mexico, Mexico; [4]Physiol. & Cell Biol., Emory Univ., Atlanta, GA

Abstract: Neuronal networks respond to challenges to their activity levels by triggering a set of homeostatic mechanisms aimed to stabilize the network firing rate. Such mechanisms are generically known as homeostatic plasticity. The most widely studied form of homeostatic plasticity is called synaptic scaling, characterized by multiplicative changes in the strength of the synaptic inputs onto a neuron. In a recent study, we demonstrated in cortical cultures that AMPAergic upscaling was triggered by reduced AMPAergic neurotransmission (receptor activation) rather than reduced spiking activity. We were able to do this by optogenetically maintaining spike rate while blocking AMPAergic transmission. In addition, we enhanced AMPA receptor activation while blocking activity with TTX by increasing mEPSC amplitude and frequency (cyclothiazide - CTZ) for 24hrs and found that this attenuated scaling. Our findings suggested that AMPAergic scaling was a mechanism of synaptic compensation rather than a homeostatic regulation of a cell's spike rate. In the current study, we intend to repeat the above experiments and test the distinct roles of spiking and neurotransmission as triggers for GABAergic scaling in mouse cortical cultures. Here we are reporting that 24hr treatment with Na+ channel blocker TTX or AMPA receptor (AMPAR) antagonist CNQX, produced a downscaling of GABAergic miniature postsynaptic current amplitude (GABA mPSC, reduced to ~53% of controls) with no significant change in GABA mPSC frequency. Simultaneous treatment of TTX and CTZ for 24hr to restore some AMPAergic neurotransmission produced a GABAergic downscaling that was no different than TTX treatment alone (reduced to ~56% of controls). These results are consistent with the idea that GABAergic synaptic scaling is triggered by reductions in spiking levels rather than AMPAergic transmission and therefore, the triggers for AMPAergic and GABAergic scaling are different. We will continue testing this possibility by maintaining spike activity optogenetically in the presence of AMPAR antagonists.

Disclosures: C.E. Gonzalez-Islas: None. **P. Buelow:** None. **P.A. Wenner:** None.

Poster

379. Homeostatic Synaptic Plasticity: Cellular and Model Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 379.05/J12

Topic: B.08. Synaptic Plasticity

Support: NIH grant MH097887

NIH grant AG055357

NIH grant NS078792

NIH grant EY012347

AHA Fellowship 16POST26560000

Title: Binding of calcium/calmodulin to PSD-95 N-terminus mediates homeostatic synaptic downscaling

Authors: *D. CHOWDHURY[1,2], M. TURNER[3], T. PATRIARCHI[2], C.-Y. CHEN[2], J. B. AMES[3], J. W. HELL[2]
[2]Dept. of Pharmacol., [3]Dept. of Chem., [1]Univ. of California Davis, Davis, CA

Abstract: The postsynaptic density scaffold protein PSD-95 is a key organizer of excitatory synapses. Abundance of glutamate receptors at the synapse, and thereby synaptic strength, is determined largely by synaptic PSD-95 abundance which, in turn, is regulated by activity. However, the mechanisms underlying the activity-induced regulation of PSD-95 synaptic localization are not clearly understood. Previous work from our lab has shown that influx of calcium following synaptic activation induces dispersal of PSD-95 from dendritic spines via binding of Ca^{2+}/Calmodulin (CaM) to the N-terminus of PSD-95 that reduces its palmitoylation (Zhang et al., *EMBO J,* 2014). The current study elucidated the role of PSD-95-CaM interaction in mediating removal of PSD-95 from synapses that accompanies homeostatic synaptic scaling down. Using peptide screening and NMR structural analysis, we identified E17 and T19 within PSD-95 N-terminus as the critical residues interacting with CaM. We generated CaM-binding-defective mutants (E17R and T19K) to test their role in PSD-95 mobilization seen in homeostatic scaling down. Using fluorescence microscopy, we observed that chronic elevation of network activity with the GABA-A receptor antagonist bicuculline reduced PSD-95 spine enrichment in cultured hippocampal neurons, but replacement of endogenous PSD-95 with either mutant blocked this effect. Moreover, each mutant blocked reduction of surface AMPA-type glutamate receptors (AMPARs) during scaling down. Further, charge inversion mutations of the interacting residues on CaM, compensating for PSD-95 mutations, restored scaling down of AMPARs. Using miniature excitatory postsynaptic current (mEPSC) analysis, the role of this interaction in regulating synaptic AMPARs in scaling down was confirmed. Overall, our findings define a specific molecular step in homeostatic synaptic scaling down. Reference: Y. Zhang, L. Matt, T. Patriarchi, Z.A. Malik, D. Chowdhury, D.K. Park, A. Renieri, J.B. Ames, J.W. Hell ‘Capping of the N-terminus of PSD-95 by calmodulin triggers its postsynaptic release’, *The EMBO Journal* 33(12): 1341-53 (2014).

Disclosures: D. Chowdhury: None. **M. Turner:** None. **T. Patriarchi:** None. **C. Chen:** None. **J.B. Ames:** None. **J.W. Hell:** None.

Poster

379. Homeostatic Synaptic Plasticity: Cellular and Model Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 379.06/K1

Topic: B.08. Synaptic Plasticity

Support: DFG CRC 1080

Title: PRG1 acts as a modulator of lipid signaling at the dendritic spine

Authors: *G. PAYRHUBER[1], W. FAN[1], A. STROH[1], R. NITSCH[2], J. VOGT[1]
[1]Univ. Med. Ctr. Mainz, Mainz, Germany; [2]Univ. Med. Ctr. Münster, Münster, Germany

Abstract: Plasticity related gene 1 (PRG1) is a transmembrane protein expressed at the postsynaptic density of excitatory neurons where it is supposed to interact with lysophosphatidic acid (LPA). Synaptic LPA binds to and activates the presynaptic LPA2 receptor leading to an increased glutamate release probability from presynaptic terminals (Trimbuch et al., 2009). Using fast 2-Photon-Microscopy live imaging and fluorescence-labeled LPA (TF-LPA), we can now show that PRG-1 plays a crucial role in LPA-uptake in postsynaptic compartments. A short puff of TF-LPA onto a dendritic spine resulted in an increase in fluorescence in the adjacent dendritic spine. This increase lasted for about 4.5 seconds in wildtype neurons, before the fluorescence level dropped down to baseline levels. However, in PRG1 KO neurons the increase in fluorescence in the spine was significantly shorter (around 2 seconds) when compared to the wildtype neurons. Using Pitstop2, a specific blocker of clathrin-dependent endocytosis, we could exclude endocytosis as a mechanism of postsynaptic LPA-uptake. Together with further pharmacological and electrophysiological characterization of this postsynaptic LPA uptake, our data suggests that PRG-1 at the postsynaptic density exerts a “pore or channel like” function critically important for synaptic LPA-uptake into postsynaptic compartments.

Disclosures: G. Payrhuber: None. **W. Fan:** None. **A. Stroh:** None. **R. Nitsch:** None. **J. Vogt:** None.

Poster

379. Homeostatic Synaptic Plasticity: Cellular and Model Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 379.07/K2

Topic: B.08. Synaptic Plasticity

Support: Whitehall Foundation Grant 2014-08-03

National Science Foundation Grant 1557792

NIH R01 Grant NS085164

Title: The long-term maintenance of homeostatic synaptic plasticity is reversible, temperature sensitive, and controlled by an intracellular calcium-handling pathway

Authors: *C. FRANK[1], T. D. JAMES[2], C. J. NEFF[3]
[1]Anat. and Cell Biol., [2]Dept. of Anat. and Cell Biol., [3]Interdisciplinary Grad. Program in Neurosci., Univ. of Iowa, Iowa City, IA

Abstract: Synapses and circuits employ homeostatic regulatory mechanisms to ensure physiologically appropriate levels of synaptic output. Improved knowledge about how forms of homeostatic synaptic plasticity (HSP) are induced and sustained throughout life should lead to an improved understanding of neurological disorders that occur when synapse stability is lost. The *Drosophila melanogaster* neuromuscular junction (NMJ) is an excellent model synapse to uncover such information. At the NMJ, genetic and pharmacological manipulations can decrease the sensitivity of postsynaptic receptors to single vesicles of neurotransmitter (quantal size). Decreased quantal size triggers retrograde, muscle-to-nerve signaling that drives increased transmitter release. In recent years, the NMJ has been key in characterizing some conserved presynaptic processes that trigger a rapid homeostatic potentiation of neurotransmitter release. However, growing evidence also demonstrates that additional factors drive the long-term maintenance of HSP. We previously described how a tyrosine kinase-based signaling system in the muscle – consisting of C-terminal Src kinase (Csk), the Neural Cell Adhesion Molecule Fasciclin II (FasII), and the Heartless (Htl) FGFR receptor – sustains the homeostatic capacity at the NMJ for long periods of developmental time. This postsynaptic signaling system activates molecules in the muscle like Target of Rapamycin (TOR) to drive HSP. We extend this prior knowledge by identifying new factors in the neuron that integrate muscle-to-nerve homeostatic signaling over long periods of developmental time. First, we identified a neuronal Phospholipase C-Beta homolog, Plc21C, as essential for the long-term maintenance of HSP. Canonically, active PLC-Beta propagates intracellular signaling by cleaving phosphatidylinositol 4,5-bisphosphase (PIP_2) into diacylglycerol (DAG) and inositol triphosphate (IP_3). These molecules then direct many cellular processes and properties, including intracellular calcium signaling and neurotransmission. Through a combination of genetic, pharmacological and electrophysiological approaches, we found that either chronic or acute perturbations in IP_3 signaling disrupt the long-term maintenance of HSP but not its short-term induction. Further, we found that this entire homeostatic maintenance process is reversible and labile at high temperatures. We propose that the IP_3 signaling pathway is essential for the long-term maintenance of homeostatic plasticity at synapses like the Drosophila NMJ, and potentially higher order synapses and circuits.

Disclosures: C. Frank: None. **T.D. James:** None. **C.J. Neff:** None.

Poster

379. Homeostatic Synaptic Plasticity: Cellular and Model Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 379.08/K3

Topic: B.09. Physiological Properties of Neurons

Support: Robert Wood Johnson Foundation

Kentucky Science and Engineering Foundation KSEF-3712-RDE-019

Personal funds

Title: Considerations in repetitive activation of light sensitive ion channels for long-term studies: Channel rhodopsin in the *Drosophila* model

Authors: *M. X. MATTINGLY, J. HIGGINS, C. HERMANNS, R. COOPER
Biol., Univ. of Kentucky, Lexington, KY

Abstract: For nervous systems to function properly, the efficacy of synapses should be finely regulated and adjustable to respond to changing circumstances and requirements. Too high or too low synaptic output results in inappropriate communication to target cells. This is most apparent during development and maturation. As the size of postsynaptic cells dramatically increases, a matched increase of neurotransmitter release and/or sensitivity of postsynaptic cell to the transmission is required. On the other hand, the nerve terminals also grow rapidly, both in size and output, and continuously show different types of remodeling to maintain proper synaptic output throughout the life of the animal. We are addressing homeostatic regulation in synaptic function at the larval Drosophila NMJ throughout development. We have examined this through optogenetic manipulation of the motor nerve terminal and muscle. The biological significance and aim of this study is to demonstrate that in controlling particular neurons or targets of neurons, over time, and throughout development, one will have a better understanding of the dynamic nature of forward and retrograde communication in regulating synaptic formation and maintenance. Optogenetics has provided a useful tool, but there are limitations in the extent of activation and inhibition which needs careful consideration. We have noted long term (minutes) unexpected effects (i.e., neuron refractory in electrical excitability) from only 10 seconds of activation of channel rhodopsin (ChR-XXL) targeting motor neurons (D42 expression). Paralysis and inability to eat are considerations for long term neural developmental studies when manipulating neurons and muscles.

Disclosures: M.X. Mattingly: None. **J. Higgins:** None. **C. Hermanns:** None. **R. Cooper:** None.

Poster

379. Homeostatic Synaptic Plasticity: Cellular and Model Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 379.09/K4

Topic: B.08. Synaptic Plasticity

Support: NIH NINDS R35NS097212

NSF GRFP

Title: Coordination of short and long-term homeostatic plasticity by an innate immune signaling pathway

Authors: *N. HARRIS[1], D. J. BRASIER[2], R. D. FETTER[1], A. TONG[1], G. W. DAVIS[1]
[1]UCSF, San Francisco, CA; [2]Carnegie Mellon Univ., Pittsburgh, PA

Abstract: It is clear that homeostatic signaling systems act throughout the central and peripheral nervous systems to stabilize the active properties of nerve and muscle. Evidence for this has accumulated by measuring how nerve and muscle respond to the persistent disruption of synaptic transmission, ion channel function or neuronal firing. In systems ranging from *Drosophila* to human, cells have been shown to restore baseline function in the continued presence of these perturbations by rebalancing ion channel expression, modifying neurotransmitter receptor trafficking and modulating neurotransmitter release. A hallmark of homeostatic plasticity is the ability to rapidly and accurately offset the magnitude of a perturbation and then sustain the homeostatic response for the duration of the perturbation, which can persist for the life of an organism. This fact raises many important questions about the nature of the homeostatic response over time. By analogy with other biological systems under homeostatic control, if a perturbation is deleterious to an organism, then an energetically costly but rapid response is warranted. If the perturbation persists, the homeostatic response can shift to an energetically less demanding, consolidated state. The nature of this transition has yet to be defined in neural homeostatic plasticity. Presynaptic homeostatic plasticity (PHP) can offset the inhibition or loss of postsynaptic neurotransmitter receptors by potentiating presynaptic vesicle release and, thereby, maintaining the gain of synaptic transmission. At the NMJ of organisms ranging from *Drosophila* to human, PHP preserves muscle excitation, thereby stabilizing movement and respiration. PHP can be rapidly induced (10 min) and stably maintained for months and even decades depending on the organism. We previously demonstrated that the innate immune receptor PGRP-LC resides at the presynaptic terminal and is essential for PHP. We now dissect the canonical signaling system downstream of PGRP-LC. We present evidence for a novel bifurcation of this signaling system in neurons. One branch of this signaling system leads to the action of a kinase, Tak1 (MAP3K), which we show is necessary for the rapid induction of PHP. The other branch signals to the NFkB-type transcription factor Relish, which is essential for the

long-term consolidation of PHP. Thus, signaling downstream of the PGRP receptor initiates, and thereby coordinates, the rapid induction and sustained, transcription-dependent consolidation of PHP. Finally, as part of our characterization, we describe the action of Tak1 on the presynaptic release mechanism.

Disclosures: **N. Harris:** None. **D.J. Brasier:** None. **R.D. Fetter:** None. **A. Tong:** None. **G.W. Davis:** None.

Poster

379. Homeostatic Synaptic Plasticity: Cellular and Model Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 379.10/K5

Topic: B.08. Synaptic Plasticity

Support: NIH NINDS R35NS097212

NIH NINDS F30NS092211

NIH GMS 5T32GM007618

Title: Post-synaptic PI3-kinase controls the set point for presynaptic homeostatic plasticity

Authors: *A. G. HAUSWIRTH, K. J. FORD, A. TONG, G. W. DAVIS
Dept. of Biochem. and Biophysics, Kavli Inst. for Fundamental Neuro, Univ. of California, San Francisco, San Francisco, CA

Abstract: Presynaptic homeostatic plasticity (PHP) is an evolutionarily conserved form of homeostatic plasticity that is expressed at synaptic connections throughout the central and peripheral nervous systems. At the neuromuscular junction (NMJ) of organisms ranging from *Drosophila* to human, PHP is induced by impaired postsynaptic neurotransmitter receptor function. The rapid induction of PHP potentiates presynaptic neurotransmitter release in seconds to minutes and precisely offsets the magnitude of postsynaptic neurotransmitter receptor inhibition. Thus, PHP can rapidly offset a postsynaptic perturbation and sustain muscle depolarization at normal baseline, set point values. Although molecular mechanisms responsible for PHP are beginning to emerge, most genes identified to date function within the presynaptic terminal to effect a change in presynaptic vesicle release. Among the few postsynaptic genes known to be required for PHP, none have been shown to be involved in the rapid induction of PHP. We sought to identify the genes necessary for the rapid induction of PHP, especially those that may act in muscle as a homeostatic sensor or as a component of the homeostatic retrograde, trans-synaptic signaling system. To do so, we performed an unbiased, RNAi-based screen of over 270 *Drosophila* kinases and phosphatases, representing 81% of the phosphatome. We identified the *Drosophila* orthologue of a class II PI3-kinase (*Pi3K68D)* as an essential post-

synaptic kinase for the rapid induction and long-term maintenance of PHP. We present cellular and genetic evidence in support of a model in which a PI(3)P membrane compartment, produced by *Pi3K68D*, creates a postsynaptic signaling platform that establishes and sustains the homeostatic set point for synaptic transmission. The concept of a 'set point' is integral to systems that are under homeostatic control, yet the cellular and molecular mechanisms that can generate a set point remain generally unknown. We propose that the set point for synaptic gain may emerge from action of a postsynaptic membrane recycling system controlled by *Pi3K68D*.

Disclosures: **A.G. Hauswirth:** None. **K.J. Ford:** None. **A. Tong:** None. **G.W. Davis:** None.

Poster

379. Homeostatic Synaptic Plasticity: Cellular and Model Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 379.11/K6

Topic: B.08. Synaptic Plasticity

Support: NIH NINDS R35NS097212

Title: Evidence for involvement of glia and epigenetic signaling in presynaptic homeostatic plasticity

Authors: *T. WANG, N. HARRIS, R. WERCBERGER, G. DAVIS
Dept. of Biochem. and Biophysics, Univ. of California, San Francisco, San Francisco, CA

Abstract: It is well-established that homeostatic signaling systems stabilize the functional properties of nerve cells and neural circuitry. Homeostasis is defined as the ability of a cell or system of cells to respond to a perturbation. If baseline neural function is precisely restored in the continued presence of a perturbation, then the process is considered to be homeostatic. The homeostatic control of presynaptic neurotransmitter release is evolutionarily conserved from *Drosophila* to human. In response to a decrease in postsynaptic neurotransmitter receptor sensitivity, a homeostatic increase in presynaptic vesicle release can precisely offset the postsynaptic perturbation and restore baseline postsynaptic depolarization. The Davis laboratory has pursued large scale forward genetic screens, using electrophysiology as the primary assay, to identify genes that are necessary for presynaptic homeostatic plasticity. To date, these studies have highlighted the action of effector proteins that are present within the presynaptic nerve terminal and, in most instances, function cell autonomously. We have uncovered evidence for the non-cell autonomous involvement of glia in presynaptic homeostatic plasticity. Synapses throughout the central and peripheral nervous systems incorporate glia into what has been termed a tripartite synapse composed of pre- and postsynaptic neuronal partners and a glial interface. We have discovered that peripheral glia are engaged by the persistent disruption of postsynaptic glutamate receptors at the *Drosophila* neuromuscular junction (NMJ). We have identified a glial-

dependent signaling system that is essential for the long-term maintenance of presynaptic homeostatic plasticity that includes the recently established function of the extracellular matrix protein Multiplexin (homologue of Collagen XVIII) and the proteolytically derived signaling molecule Endostatin. This signaling system includes evidence of epigenetic transcriptional control that may be directly involved in the glial-dependent, persistent regulation of presynaptic neurotransmitter release during homeostatic plasticity.

Disclosures: T. Wang: None. **N. Harris:** None. **R. Wercberger:** None. **G. Davis:** None.

Poster

379. Homeostatic Synaptic Plasticity: Cellular and Model Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 379.12/K7

Topic: B.08. Synaptic Plasticity

Support: NIH NINDS R35NS097212

Title: Genetic intersection of autism gene mutations and homeostatic plasticity

Authors: *O. GENC, G. W. DAVIS
UCSF, San Francisco, CA

Abstract: Autism spectrum disorders (ASD) are considered to have strong genetic basis and a large number of genes have been identified that confer high risk for ASD in humans. Yet, it remains unknown how genetic risk translates into the phenotypic severity of ASD in a given individual. We hypothesize that homeostatic plasticity represents a physiological buffer that limits the phenotypic severity of an ASD-associated gene mutation. An extension of this hypothesis is that an ASD mutation could genetically interact with one or more second site, heterozygous mutations in the genetic background of an organism. In rare instances, if these genetic interactions cause homeostatic plasticity to fail, then the buffering effect of homeostatic plasticity would be limited and the phenotypic severity of the ASD mutation would be enhanced. To test this hypothesis, we are systematically combining heterozygous ASD mutations with deficiency chromosomes that uncover defined regions of the Drosophila genome. In each genetic combination, we assess the expression of presynaptic homeostatic potentiation (PHP) by direct quantification of synaptic transmission. The results of this large-scale, functional genetic screening effort will be presented, including the identification of gene-specific mutations that functionally interact with, and enhance, the phenotype of heterozygous ASD mutations. Based on the results of this screen, we propose that failure of presynaptic homeostatic plasticity may be a common underlying pathophysiology relevant to the phenotypic penetrance of rare de novo mutations that confer high risk for ASD in human.

Disclosures: **O. Genc:** None. **G.W. Davis:** None.

Poster

379. Homeostatic Synaptic Plasticity: Cellular and Model Systems

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 379.13/K8

Topic: B.08. Synaptic Plasticity

Title: Circadian genes, sleep, and alcohol in *Drosophila*

Authors: *A. GHEZZI[1], M. E. RAMIREZ[1], J. L. AGOSTO[1], N. S. ATKINSON[2]
[1]Dept. of Biol., Univ. of Puerto Rico, Rio Piedras, San Juan, PR; [2]Dept. of Neurosci., The Univ. of Texas at Austin, Austin, TX

Abstract: Exposure to alcohol is known to trigger homeostatic adaptations in the brain that lead to the development of drug tolerance and dependence. These adaptations are also believed to be the root of a collection of sleep disturbances that often manifest during the development of alcoholism. Because both, alcohol dependence and sleep modulation are under homeostatic regulation, we hypothesize that these processes share common mechanisms. Here we use a Drosophila model system to test this hypothesis. We find that in Drosophila, acute alcohol exposure causes disturbances in sleep patterns that resemble those described in mammals. These disturbances include an increase in total sleep duration, decrease sleep latency, as well as an increased number of sleep episode per day (fragmented sleep). Furthermore we show that many of the genes implicated in the neural adaptations behind alcohol tolerance, have also been implicated in the regulation of sleep cycles. Using a novel genomic approach that exploits the analysis of epigenetic modifications and transcription factor binding we identify a network of alcohol-induced genes that is directly controlled by the circadian clock genes Clock and cycle. These results support our hypothesis that sleep and alcohol neuroadaptation share a common regulatory mechanism, and brings us closer to understanding the interaction between these two homeostatic processes.

Disclosures: **A. Ghezzi:** None. **M.E. Ramirez:** None. **J.L. Agosto:** None. **N.S. Atkinson:** None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.01/K9

Topic: B.09. Physiological Properties of Neurons

Support: WaveFrontImag, program number ANR-14-CE17-0006-01

Labex Ion Channels Science and Therapeutics, program number ANR-11-LABX-0015

Federation pour la recherché sur le Cerveau (FRC) grant Espoir en tête (in partnership with Rotary France)

Title: Membrane depolarisation, inactivation of A-type K^+ channels and transient saturation of Ca^{2+} buffers, mediated by mGluR1, synergistically control the spatial segregation of climbing fibre signalling in cerebellar Purkinje neurons

Authors: *K. AIT OUARES, L. FILIPIS, M. CANEPARI
Liphy, Saint Martin D Héres, France

Abstract: Several forms of associative synaptic plasticity depend on the coincidence between local synaptic activation and transient widespread membrane depolarisation. In cerebellar Purkinje neurons, the transient depolarisation that regulates parallel fibre (PF) synaptic plasticity is provided by the climbing fibre (CF), but the precise machinery permitting this unspecific dendritic depolarisation to produce a Ca^{2+} signal localised in activated synaptic regions has been elusive. Here we used a series of advanced optical techniques to reveal in detail the sequence of events following activation of PF synapses and leading to a local large Ca^{2+} signal associated with the CF depolarisation. Using ultrafast Ca^{2+} imaging with the low affinity indicator Oregon Green BAPTA-5N, computational extraction of Ca^{2+} currents and membrane potential imaging, we demonstrate that the nonlinear amplification of the CF-associated Ca^{2+} signal is determined by the synergy of several mechanisms. Burst activation of PF inputs initially produces sustained dendritic depolarisation and Ca^{2+} entry via voltage gated Ca^{2+} channels. With a delay of a few tens of milliseconds, the released glutamate, activates type-1 metabotropic glutamate receptors (mGluR1s) enhancing inactivation of A-type K^+ channels in combination with dendritic depolarisation. The fast cytosolic Ca^{2+} elevation, initially via T-type Ca^{2+} channels Ca^{2+} influx, and later via mGluR1-activated TRP3 channels, transiently saturates the endogenous Ca^{2+} buffers. Thus, when a CF excitatory postsynaptic potential (EPSP) occurs at the peak of mGluR1 activation, the inactivation of A-type K^+ channels leads to activation of P/Q-type Ca^{2+} channels and the previous Ca^{2+} binding to endogenous buffers amplifies the fast Ca^{2+} elevation associated the CF-EPSP locally in the small dendritic region adjacent to the activated mGluR1s. In this

way, the non-selective CF depolarisation can produce a triggering signal only in activated PF synaptic regions.

Disclosures: K. Ait Ouares: None. **L. Filipis:** None. **M. Canepari:** None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.02/K10

Topic: B.09. Physiological Properties of Neurons

Support: CIHR grant MT-12682

CIHR CGS award

Title: Action potential-induced calcium responses actively backpropagate in spinal cord lamina I neurons

Authors: *E. K. HARDING[1,2], M. W. SALTER[1,2]
[1]Hosp. For Sick Children, Toronto, ON, Canada; [2]Physiol., Univ. of Toronto, Toronto, ON, Canada

Abstract: Spinal cord lamina I neurons function as a hub of nociception, and exhibit hyperexcitability in chronic pain models. Calcium is a second messenger critical to the induction of many cellular processes, including long-term potentiation (LTP) in many types of neurons. With LTP and lamina I neuron hyperexcitability sharing many characteristics, it is possible that calcium entry could be a critical mechanism underlying hyperexcitability. However, the function of calcium in lamina I neurons is poorly understood. To address this, we combined two-photon calcium imaging with whole cell electrophysiology. We made current-clamp recordings from lamina I neurons in spinal cord slices, loading the calcium indicator OGB-1 and control fluorophore AF-594 via the patch pipette. Neuronal calcium levels before, during, and after action potential (AP) generation were quantified as ΔG/R (change in OGB-1 intensity as compared to AF-594). Recordings were made from over 180 lamina I neurons. In 99% of neurons a single AP was sufficient to increase ΔG/R in the somatic cytosol (ΔG/R=0.11, n=179 cells), nucleus (ΔG/R=0.047, n=179 cells), dendrites (ΔG/R=0.15, n=179 cells), and dendritic spines (ΔG/R=0.091, n=20 spines). These calcium responses were ablated by the VGCC blocker cadmium. Responses did not degrade along dendrites from 20µm (ΔG/R=0.10, n=6 dendrites) to 160µm (ΔG/R=0.13, n=6 dendrites) away from the soma, indicating dependence on VGCCs and actively backpropagating action potentials (bAPs). We found no differences when grouping calcium responses by cellular morphology (fusiform, multipolar, pyramidal) or firing-type (tonic, phasic, single spike, delayed onset). We compared lamina I neuron calcium responses to

hippocampal CA1 neurons, and found that lamina I responses (cytosol ΔG/R=0.063, decay rate=1.73s; nucleus ΔG/R=0.022, decay rate=6.84s; dendrite ΔG/R=0.067, decay rate=1.48s, n=4 cells) had a decreased amplitude and decay rate compared to hippocampal neurons (cytosol ΔG/R=0.18, decay rate=0.49s; nucleus ΔG/R=0.055, decay rate=3.65s ; dendrite ΔG/R=0.13, decay rate=0.35s, n=4 cells). This is suggestive of increased immobile calcium buffer in lamina I neurons. Together, these findings strongly suggest that all lamina I neurons regardless of cell type produce active bAPs that invade the entire dendritic arbour. The presence of active bAPs gives lamina I neurons the capacity to readily undergo both short and long-term potentiation through temporal summation and spike-timing dependent plasticity (STDP) respectively.

Disclosures: E.K. Harding: None. **M.W. Salter:** None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.03/K11

Topic: B.09. Physiological Properties of Neurons

Support: NCBS/TIFR

DST India grant SR/CSI/66/2013

Title: Multiscale model of feedback and activity modulation in stimulus sequence recognition by chemical signaling in dendrites

Authors: *U. S. BHALLA
Natl. Ctr. For Biol. Sci., Bangalore, India

Abstract: Introduction:
We propose that the key neuronal computation of sequence discrimination occurs through chemical signaling in dendrites on length scales of 5-30 microns, and on behavioural timescales of ~1 second per stimlus. Synaptic input arriving on a series of closely spaced (~2 microns) synapses, in sequential order, is discriminated from scrambled input. In these systems, synaptic input triggers electrical as well as chemical activity through calcium influx. In turn, signaling cascades modulate electrical excitability through channel phosphorylation and receptor insertion. We investigated the interplay between these scales of signaling in the context of sequential vs. scrambled synaptic input, with different kinds of background activity and neuromodulation.
Methods:
We performed detailed multiscale simulations using experimental neuronal morphologies from rat hippocampal neurons (e.g., NeuroMorpho.org accession VHC-neuron.CNG.swc), embroidered with ~5500 dendritic spines. We incorporated voltage-gated ion channels and

calcium dynamics all over the cell, and added AMPA and NMDA receptors on all spines. We also incorporated a reaction- diffusion system based on MAPK signaling throughout the dendrites, and calcium signaling including buffering by calmodulin, in all spines and dendrites. The model was simulated using stochastic calculations in the spines, and deterministic calculations in the dendrites. The computations were done using the MOOSE simulator (moose.ncbs.res.in) on Linux workstations and supercomputers.
Results:
We demonstrate that a single neuron can discriminate thousands of input sequences in parallel over its dendritic tree, responding only when at least one of these sequences arrives on successive synapses in the correct order. We investigate how the preferred timing for sequence recognition can be modulated by stimulus amplitude, background activity, and neuromodulators. We find that the feedback between electrical and chemical signaling leads to runaway activation for some parameter domains, but there is a substantial domain where there is good sequence selectivity followed by modulation of cellular firing activity.

Disclosures: U.S. Bhalla: None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.04/K12

Topic: B.09. Physiological Properties of Neurons

Support: Wellcome Trust

European Research Council

Marie-Skłodowska Curie Postdoctoral Fellowship

Title: Fast 3D imaging of dendritic activity in pyramidal cells of visual cortex

Authors: *T. J. YOUNTS, C. BARAGLI, A. SILVER
Neuroscience, Physiol. and Pharmacol., Univ. Col. London, London, United Kingdom

Abstract: How sensory information is represented in dendrites in vivo is poorly understood because it is difficult to measure activity within morphologically complex dendrites. We used a custom built 3D two-photon acousto-optic lens microscope to measure dendritic activity in pyramidal cells in mouse primary visual cortex. Pyramidal neurons were sparsely co-labelled with the genetically-encoded Ca^{2+} indicator GCamp6f (to measure activity) and with the red fluorophore TdTomato (to detect and later correct for movement). Random access point measurements were collected at 80-120 Hz from hundreds of dendritic locations distributed in 3D. Patterns of activity were characterized in layer 2/3 and layer 5 pyramidal cells during

presentation of square wave gratings in animals that were anaesthetized, awake and stationary, or running on a wheel. Layer 2/3 neurons exhibited Ca^{2+} transients that were mainly local. Somatic Ca^{2+} transients were rarely accompanied by global events in the full dendritic tuft. Dendritic activity was affected little by anaesthesia and locomotion. In the tuft of layer 5 neurons, Ca^{2+} transients tended to be either localized to few dendritic branches or to occur in the full-dendritic tuft (and soma when measurable). Dendrites showed little response to visual stimuli, but they were strongly modulated by the state of the animal: anaesthesia suppressed activity while locomotion enhanced activity when compared to rest. To investigate the mechanisms underlying these dendritic activity patterns observed in vivo (i.e. global vs. local), we made whole cell patch clamp recordings from layer 2/3 and layer 5 pyramidal cells in acute visual cortical slices. 3D dendritic imaging of GCaMP6f or the synthetic Ca^{2+} fluorescence indicator Cal520 revealed that single and bursts of action potentials produce Ca^{2+} transients that backpropagate and invade the distal dendritic tuft of both cell types in young (<P30) mice, but responses to single spikes were not detected in the tuft of layer 5 cells in adult mice (>P60). These data indicate that layer 2/3 and layer 5 pyramidal neurons exhibit different patterns of activation in their dendritic trees. Our in vivo results are consistent with the view that layer 2/3 neurons are predominantly driven by bottom-up feedforward visual inputs, while top-down inputs conveying information about the animal's state are effective in activating the apical dendrites of layer 5 cells.

Disclosures: **T.J. Younts:** None. **C. Baragli:** None. **A. Silver:** None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.05/L1

Topic: B.09. Physiological Properties of Neurons

Support: NSERC RGPIN 326804-12

NIH R01 MH094839

NIH R01 NS084473

Title: Optimal signal transfer from dendrite to soma in mouse L5 pyramidal neurons

Authors: *E. P. COOK[1], R. A. GRAY[2], D. JOHNSTON[2], B. E. KALMBACH[3]
[1]McGill Univ., Montreal, QC, Canada; [2]Ctr. for Learning and Memory, The Univ. of Texas At Austin, Austin, TX; [3]Allen Inst. for Brain Sci., Seattle, WA

Abstract: What is the optimal dendritic input for producing either somatic depolarizations or somatic action potentials (APs)? We addressed this question using previously published simultaneous dendrite and soma electrophysiological recordings from PFC mouse L5 pyramidal

neurons stimulated with white-noise dendritic current injections. Specifically, we focused on how the energy contained in the dendritic input signal is transformed into somatic depolarizations or action potentials. An optimal input signal is defined as one that produces an equivalent somatic response with the least amount of energy. Using a white-noise dendritic stimulus is an advantageous approach because it allowed us to easily identify the shape of the optimal input that maximized the energy transfer from the dendrites to the soma. For each neuron (N = 16), we identified two unique optimal input signals: First, we estimated the linear transfer function (Gdep) that converted input current into subthreshold somatic depolarization. Second, we computed the average noise stimulus that preceded each AP (the spike-triggered average or Gap). These two functions describe the stimulus shape that results in optimal energy transfer from the dendrite to either somatic depolarization (Gdep) or AP (Gap). We found that Gdep and Gap had very different shapes. Although both Gdep and Gap showed theta-band resonance, Gdep attenuated input energy at frequencies above ~10 Hz while Gap contained substantial energy above 100 Hz. Importantly, the signal energy required to produce subthreshold somatic depolarizations was highly dependent on dendritic location (range of 50 to 380 μm), while the amount of signal energy preceding an AP was not dependent on dendritic location. Thus, the optimal input signal for producing a somatic AP contained high frequencies that would normally be eliminated by the passive filtering properties of dendrites. Although this result seems contradictory, a previously described functional model of the dendritic nonlinearity and somatic integrator reproduced our experimental observations. Our results show that by adding energy to the dendritic input, the dendritic nonlinearities normalize dendritic location and shape the optimal input signal that maximally transfers energy to the soma.

Disclosures: **E.P. Cook:** None. **R.A. Gray:** None. **D. Johnston:** None. **B.E. Kalmbach:** None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.06/L2

Topic: B.09. Physiological Properties of Neurons

Support: NIH grant MH065339

NSF grant DMS-1120952

Title: Active dendrites broaden membrane bandpass filtering and decrease temporal synchrony in a looming-sensitive neuron

Authors: *R. B. DEWELL[1], F. GABBIANI[2]
[1]Dept of Neurosci., Baylor Col. of Med., Houston, TX; [2]Baylor Col. Med., Houston, TX

Abstract: Filtering and integration of synaptic inputs is critical for neural computations. A well-studied looming-sensitive neuron in the locust that integrates ~15,000 retinotopically organized excitatory synaptic inputs has long served as a model for understanding the neural computations underlying collision avoidance behaviors. This neuron, the lobula giant movement detector (LGMD) is precisely tuned for the pattern of retinal activation generated by an object approaching on a collision course. Previous work has shown that active dendritic channels, including Ih, increase the LGMD's neuronal and the animal's behavioral specificity through improved discrimination of the resulting spatiotemporal synaptic patterns. The LGMD contains prominent h and M currents, both of which can produce membrane resonance and bandpass filtering in neurons. Additionally, Ih has also been shown to increase the detection of temporally synchronous inputs within some cortical neurons by reducing the phase lag of signals as they propagate from distal dendrites toward the site of spike initiation. Surprisingly, we found that in the LGMD these currents combine to make the membrane less resonant and more broadband. Ih also increased the phase lag of dendritic signals propagating towards the spike initiation zone, which could increase the ability of the LGMD to discriminate between spatiotemporal patterns of synaptic inputs. As the spatiotemporal pattern of synaptic inputs into the LGMD is important both for the computations carried out within the neuron and the animal's escape behavior, this neuron represents an attractive model to investigate the influence of active conductances on the resonance and filtering of synaptic inputs.

Disclosures: R.B. Dewell: None. **F. Gabbiani:** None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.07/L3

Topic: B.09. Physiological Properties of Neurons

Support: NSF Grant IOS-1516235

NSF Grant DMS-1547394

Title: Finding thresholds in active dendrites

Authors: *W. L. KATH
Applied Mathematics, Northwestern Univ., Evanston, IL

Abstract: The integration of synaptic inputs in a neuron can be nonlinear not just at the axon, but also locally in the dendrites if they contain active voltage-gated ion channels. For example, CA1 pyramidal neurons have high densities of sodium and potassium and currents in their dendrites, and the densities can vary substantially with location in the arbor. When combined

with diameter changes at branch points in the dendritic tree, such nonlinearities can lead to compartmentalized responses to inputs: each branch can act as an individual nonlinear unit in which action-potential-like events known as dendritic spikes occur.
To fully comprehend how pyramidal neurons integrate their inputs, it is therefore necessary to understand the conditions under which combined synaptic inputs initiate dendritic spikes, and how and when these spikes interact with other inputs to generate a somatic action potential.
In contrast to single cell neuron models, however, the dividing line between an active and non-active event in a full compartmental model is a steady-state, unstable, spatially-varying solution. These properties (in particular, instability and spatially variation) make finding such dividing lines and the critical synaptic conductance(s) leading to them troublesome.
The steady-state solution often has only a single unstable eigenvalue, however, leading to several methods to find the dividing line between super- and sub-threshold events. First of all, a projective shooting method that focuses on dynamics in the unstable direction can be used. Second, one can employ a Recursive Projection Method, a functional iteration devised to find steady-states with a finite number of unstable eigenmodes. Finally, Newton's method can be used. The pros and cons for each of these methods will be discussed. In addition, examples will be given showing how the method determines changes in the threshold for dendritic spikes due to variations in branch geometry or ion channel densities.

Disclosures: W.L. Kath: None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.08/L4

Topic: B.09. Physiological Properties of Neurons

Support: NIH Grant MH104602

Title: The role of dendritic spikes in inducing timing dependent plasticity in CA1 pyramidal neurons

Authors: *T. BOCK[1], S. A. SIEGELBAUM[2]
[1]Dept. of Neurosci., Columbia Univ., New York, NY; [2]Dept of Neurosci., Columbia Univ. Coll P & S, New York, NY

Abstract: Input time dependent plasticity (ITDP), a powerful form of heterosynaptic plasticity in CA1 pyramidal neurons (PNs), has been implicated in contextual learning and object memory storage. ITDP requires precisely timed pairing of perforant pathway (PP) input from entorhinal cortex (EC) to the distal dendrites of CA1 PNs with excitatory input to CA1 proximal dendrites from the CA3 Schaffer collaterals (SC) after a 20 ms delay, leading to a 2-3 fold enhancement of

the SC-CA1 PSP, largely caused by CB1 receptor-mediated LTD of feedforward inhibition from CCK-positive interneurons. Induction of ITDP is associated with the firing of dendritic calcium spikes in the dendrites of CA1 PNs during paired stimulation, although these spikes have little impact on somatic voltage. Here we report that dendritic spikes are both necessary and - in conjunction with SC input alone - sufficient to induce ITDP. We paired direct current injection into distal apical dendrites through a dendritic patch pipette with synaptic stimulation of SC inputs. This pairing protocol was sufficient to induce a 2-fold increase in the SC PSP that was blocked by bath application of the CB1 cannabinoid receptor blocker AM251 during ITDP induction and was occluded by blocking GABA receptors, demonstrating that this plasticity shared a similar expression mechanism with synaptically induced ITDP. Furthermore, dendritic spikes were necessary for ITDP induction. Closed loop negative current injection into the distal dendrite was used to prevent spike firing without altering subthreshold EPSPs. Spike inhibition in this manner prevented the induction of ITDP during paired PP and SC stimulation. Thus we conclude that dendritic spikes are both sufficient and necessary to induce ITDP in CA1 pyramidal neurons. The main impact of the EC-CA1 pathway on ITDP induction is likely through the combined effects of distal dendritic depolarization combined with disinhibition of the distal apical dendrite through long-range inhibitory projections onto dendrite-targeting CCK interneurons. This form of precisely timed disinhibition facilitates the generation of dendritic spikes in the distal apical dendrite during SC stimulation, which in turn are the main drivers of ITDP induction.

Disclosures: **T. Bock:** None. **S.A. Siegelbaum:** None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.09/L5

Topic: B.09. Physiological Properties of Neurons

Support: BMBF No. 01GQ1406 – Bernstein Award 2013 to H.C.

BMBF No. 01GQ1203A to P.J.

DFG CRC1080 to T.D.

DFG CU-217-2 to H.C.

IMPRS Neural Circuits to F.Z.H.

Title: Neuronal responses to distributed synaptic inputs are generally independent of dendritic length and branching structure

Authors: *H. CUNTZ[1,2], A. D. BIRD[1,2], M. BEINING[1,2,3], F. Z. HOFFMANN[1,2], S. PLATSCHEK[3], T. DELLER[3], P. JEDLICKA[3]
[1]Ernst Strüngmann Inst. (ESI), Frankfurt, Germany; [2]Frankfurt Inst. for Advanced Studies, Frankfurt, Germany; [3]Inst. of Clin. Neuroanatomy, Goethe Univ., Frankfurt, Germany

Abstract: Reducing neuronal size results in less cell membrane and therefore lower input conductance. Consequently, smaller neurons show an increase in excitability and an altered input-output function in response to current injections in the soma [1, 2]. However, the general consequences of this relation for responses to distributed synaptic inputs have yet to be studied in detail. Using analytical cable theory we find that distributed synaptic inputs effectively encounter an apparent input conductance corresponding to the collapsed membrane leak of the entire dendrite onto the soma. As a consequence more synaptic currents are precisely compensated by more dendritic leak and the voltage response at the soma is independent of dendritic length, complexity or topology. Assuming a given synaptic density, voltage responses only depend on the average diameter and the specific membrane conductance in the dendrite. We show in passive and active model simulations that this relation holds for realistic morphologies and is reflected in invariant numbers of spikes for a given synaptic input frequency. In conclusion, principles of neural architecture seem to provide for neural input-output functions that are generally independent of dendrite length and shape within the range of most realistic biological neurons. This represents a homeostatic dendritic input-output relation that dendritic non-linearities and synaptic plasticity can freely modulate.
References:
[1] Mainen ZF, Sejnowski TJ (1996) Influence of dendritic structure on firing pattern in model neocortical neurons. Nature 382:363-366.
[2] Stuart GJ, Spruston N (2015) Dendritic integration: 60 years of progress. Nat Neurosci 18:1713-1721.

Disclosures: H. Cuntz: None. **A.D. Bird:** None. **M. Beining:** None. **F.Z. Hoffmann:** None. **S. Platschek:** None. **T. Deller:** None. **P. Jedlicka:** None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.10/L6

Topic: B.09. Physiological Properties of Neurons

Support: Wellcome Trust 090915

NKFIH ERC_HU_15 119023

Title: Input pattern dependent cooperative synaptic plasticity in dendrites of hippocampal CA1 pyramidal cells

Authors: A. MAGO, J. WEBER, *J. K. MAKARA
Inst. of Exptl. Med., Budapest, Hungary

Abstract: Synaptic inputs arrive to neurons in various patterns, defined by their number, synchrony and spatial distribution in the dendritic tree. Depending on the input pattern, dendrites may integrate synaptic voltage signals either linearly, sublinearly or supralinearly (the latter prominently by local dendritic spikes), allowing the cell to specifically process and respond to different synapse patterns. It is less clear how long-term plasticity of synaptic inputs depend on their pattern and dendritic integration mode. We have recently shown that, in distal segments of perisomatic dendrites of CA1 pyramidal cells, coactivation of a few spatially clustered synapses can lead to their local interaction that produces linear voltage integration but supralinearly amplified synaptic NMDA receptor mediated Ca^{2+} signals. Furthermore, repetitive synchronous activation of four clustered distal inputs can induce NMDAR dependent long-term potentiation (LTP) of the activated synapses (Weber et al., 2016 Nature Comm.). Here we characterized the pattern requirements of this plasticity in details, and examined how cooperative plasticity of different input patterns depends on the voltage integration mode by the dendrite. We used two-photon imaging and multisite glutamate uncaging combined with whole cell current clamp recordings to evoke linear or supralinear dendritic responses by different spatial input patterns at various dendritic locations. We found that cooperative LTP with linear voltage integration could be evoked by 3-4 clustered synapses, but only at distal and not at proximal dendritic sites. On the other hand, spatially distributed distal synapses evoked LTP only if they generated dendritic spikes. At proximally located inputs, LTP could be only evoked by patterns that generated dendritic, or backpropagating somatic spikes, but not by subthreshold synapse clusters. These results indicate that the induction mode of cooperative LTP follows location dependent rules in dendrites, and suggests a gradual switch from global to more local mechanisms of LTP along the proximodistal axis of individual thin branches. The diversity of pattern dependent cooperative synaptic plasticity rules may enrich the versatility of hippocampal information storage.

Disclosures: A. Mago: None. **J. Weber:** None. **J.K. Makara:** None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.11/L7

Topic: B.09. Physiological Properties of Neurons

Support: European Union Horizon 2020 Research and Innovation Programme (under grant agreement numbers 72070/HBP SGA1 and 670118/ERC ActiveCortex)

European Union's Seventh Framework Programme for research, technological development and demonstration under grant agreement no: Health-F2-602531-2013 (DESIRE)

Deutsche Forschungsgemeinschaft (EXC NeuroCure, LA 3442/3-1, SPP1665, SFB1078 B2)

NeuroCure Cluster of Excellence REF: DFG, EXC 257

Title: Back- and forward- propagating action potentials in dendrites of human layer 3 cortical pyramidal neurons

Authors: *A. GIDON[1], T. A. ZOLNIK[2,1], F. BOLDUAN[4], P. FIDZINSKI[3], M. HOLTKAMP[3], I. VIDA[4,5], M. E. LARKUM[1]
[1]Inst. for Biol., Humboldt Univ. zu Berlin, Berlin, Germany; [3]Epilepsy-Center Berlin-Brandenburg Dept. of Neurol., [2]Charité - Universitätsmedizin Berlin, Berlin, Germany; [4]Inst. for Integrative Neuroanatomy, Charité - Universitätsmedizin Berlin, Berlin, Germany; [5]NeuroCure Cluster, Charité - Universitätsmedizin Berlin, Berlin, Germany

Abstract: The back propagating action potential (bAP) is considered a global dendritic signal that broadcasts the neuron's spike output to the synaptic input sites on its dendrite. Thus, it is thought to have a key role in Hebbian plasticity and synaptic integration. However, our understanding of bAPs is derived mostly from rodent neurons, thus it is unknown to what extent the properties of rodents' bAPs apply in other species. Here, for the first time, we used somato-dendritic patch clamp and two-photon calcium imaging to record the bAP in human layer 3 neocortical pyramidal cell (L3PC) apical dendrites and spines obtained from resected brain tissue of epileptic patients. We found that the bAP in human L3PC dendrites severely attenuates; although L3PC dendrites often extend up to more than 1 mm, the bAP did not reach beyond 500 μm from the soma. However, high frequency bursts (200 Hz) invaded significantly farther into the dendrite. Furthermore, in some cases, sustained depolarization of the dendrite caused repetitive dendritic spikes that propagated forward to the soma - i.e., forward-propagating dendritic spikes (fAPs). Often, fAPs were capable of reliably triggering somatic spikes. In conclusion, this study shows that AP propagation in the apical dendrites of human L3PCs is bidirectional, a result that has important implications for communication between soma and dendrites and plasticity rules in the human brain.

Disclosures: **T.A. Zolnik:** None. **F. Bolduan:** None. **P. Fidzinski:** None. **M. Holtkamp:** None. **I. Vida:** None. **M.E. Larkum:** None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.12/L8

Topic: B.09. Physiological Properties of Neurons

Title: Cellular mechanisms for multimodal integration in retrosplenial cortex

Authors: *M.-S. VAN DER GOES[1], M. LAFOURCADE[2,1], D. BARNAGIAN[2,1], M. T. HARNETT[1,2]
[1]Dept. of Brain and Cognitive Sci., MIT, Cambridge, MA; [2]MIT, McGovern Inst. for Brain Res., Cambridge, MA

Abstract: How association cortices in the mammalian brain perform multimodal integration of sensory, contextual, and motor information remains unknown. We hypothesized that this process could occur at the level of individual cortical pyramidal neurons as a result of active dendritic mechanisms controlling the interaction of compartmentalized synaptic input streams. We tested this hypothesis in retrosplenial cortex (RSC), a cortical association area engaged in spatial navigation, specifically the integration of world-centered (i.e. directional) and egocentric signals (i.e. visual). First, we used monosynaptic retrograde tracing to identify a variety of brain regions that directly targeted specific neuron classes in RSC. These regions include primary visual (V1) areas, secondary visual (V2) and motor (M2) areas, and cingulate cortices, as well as thalamic and subcortical nuclei processing head orientation, suggesting that multiple streams of information converge onto single cells. Channelrhodopsin-assisted subcellular input mapping was then used to reveal the synaptic connectivity logic of a subset of identified input regions (V1, anterior and lateral dorsal thalamic nuclei, and M2). We observed discrete and spatially organized dendritic input targeting specific RSC cell classes, suggesting the presence of highly-structured, dedicated channels for information processing at the cellular level. Finally, we employed a combination of patch-clamp electrophysiology with two-photon Ca^{2+} imaging and glutamate uncaging to reveal multiple, distinct dendritic integration mechanisms segregated to particular dendritic domains in RSC pyramidal neurons potentially congruent with observed input connectivity maps. Specifically, we found that basal dendrites exhibit canonical NMDA receptor-mediated supralinear input-output processing, while proximal apical dendrites integrate in a surprisingly linear manner until action potential initiation. We conclude that the functional and anatomical organization of excitatory synaptic inputs and the presence of diverse dendritic decoding motifs provide a cellular substrate for multimodal integration in RSC. Future experiments in head-fixed, task-performing mice will test how subcellular connectivity and dendritic processing operations interact with pre- and post-synaptic circuit-level dynamics to generate the computations in RSC that guide spatial behavior.

Disclosures: **M. Van Der Goes:** None. **M. Lafourcade:** None. **D. Barnagian:** None. **M.T. Harnett:** None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.13/L9

Topic: B.09. Physiological Properties of Neurons

Title: Hierarchical input integration at glutamatergic synapses of the nucleus accumbens

Authors: ***M. DEROCHE**[1,2], F. MICHEL[1], O. MANZONI[1]
[1]Inserm Unit 901, Inmed, Marseille, France; [2]Aix-Marseille Univ., Marseille, France

Abstract: The nucleus accumbens located at the center of mesocorticolimbic system regulates emotional and reward-related behavior. We have performed a comprehensive morphological and electrophysiological description of the morpho-functional properties of identified cell types (i.e. D1R or D2R-expressing GABAergic medium spiny neurons) in the nucleus accumbens mice core. To this aim, we have systematically audited the tri-dimensional organization of dendritic trees, intrinsic neuronal properties, synaptic transmission and plasticity at optogenetically-disambiguated synapses on identified neuronal subtypes (i.e. D_1R+ or D_1R- accumbens medium spiny neurons) ex-vivo. Finally, to establish a link between structural organization and functional output, we visualized the whole brain connectivity map of the excitatory afferents to identified accumbens neurons with clarifying approaches. Our multiparametric dataset reveals the precise hierarchical organization of the excitatory inputs to accumbens medium spiny neurons.

Disclosures: **M. Deroche:** None. **F. Michel:** None. **O. Manzoni:** None.

Poster

380. Dendritic Properties and Activity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 380.14/L10

Topic: B.09. Physiological Properties of Neurons

Support: NIH Brain Initiative Grant R01 EB022903 01

Title: Dendritic plateau generation model in cortical pyramidal neurons: A link to cortical ensembles

Authors: *P. GAO[1], J. W. GRAHAM[2], S. ANGULO[2], S. DURA-BERNAL[2], W. W. LYTTON[2], S. D. ANTIC[1]
[1]Univ. of Connecticut Hlth. Ctr., Farmington, CT; [2]Physiol. & Pharmacol., SUNY Downstate, Brooklyn, NY

Abstract: Synaptic and glutamatergic inputs to the basal dendrites of cortical pyramidal neurons activate NMDA and AMPA receptors which can bring the dendrites into a long-lasting depolarized state which can spread into the soma. The resulting plateau potentials increase the excitability of the neuron by bringing membrane potential closer to threshold and reducing membrane time constant.
In such an "activated" state, the pyramidal cells can respond to synaptic inputs more quickly and easily, facilitating synchronization of firing. We hypothesize that a multiple embedding of neuronal ensembles would allow for the binding of multimodal inputs into coherent object perception based on binding-by-synchrony theory, while simultaneously allowing information transfer via temporal coding. The first level of embedding is the activated neurons among their larger populations, and the second level consists of the activated neurons which are firing synchronously. Synchronized temporal coding within an area may provide the substrate for a broad distributed ensemble across areas that would allow binding-by-synchrony.
We are developing two data-driven models of cortical pyramidal neurons in NetPyNE for the NEURON simulator: 1) a morphologically-simplified model with seven compartments and 2) a morphologically-accurate model reconstructed from a prefrontal cortex Layer 5 pyramidal neuron. Electrophysiological data is available from dendrites and soma simultaneously with glutamate-uncaging experiments utilizing voltage-sensitive dyes in dendrites and whole-cell patch recording in the soma.
Model cell properties were fitted to mouse M1 current-clamp results using PRAXIS and Evolutionary Multi-objective Optimization (EMO) algorithms. Dendritic active current properties were tuned to match experimentally observed back-propagating action potentials. An AMPA receptor model generates the fast-response to glutamate stimulation while an NMDA receptor model accounts for the extended plateau activation.
The AMPA/NMDA receptor properties were explored to determine their effect on dendritic and somatic plateaus. The receptors were modeled both directly on a basal dendrite and on spines. Spine properties, glutamate stimulation properties, and stimulation location were all explored and quantified. These models are being used to develop cortical meso-scale network models to develop and explore EEE theory.

Disclosures: P. Gao: None. **J.W. Graham:** None. **S. Angulo:** None. **S. Dura-Bernal:** None. **W.W. Lytton:** None. **S.D. Antic:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.01/M1

Topic: B.10. Network Interactions

Support: NEI Grant 1R01EY026156-01A1

Title: Pinging the network - Causal modulation of pairwise correlations using optogenetics

Authors: *A. R. ANDREI, S. POJOGA, R. JANZ, V. DRAGOI
Neurobio. & Anat., Univ. of Texas Hlth. Sci. Ctr. At Houston, Houston, TX

Abstract: Noise correlations, or correlated variability, represent a well-studied measure of cortical networks that have been shown to vary across many conditions, such as cortical layers, attentional and arousal states, or task demands. Further, pairwise correlations have been proposed to limit the amount of information encoded by a network. To test whether changes in correlations impact information coding, causal manipulations in the absence of concomitant firing rate changes are necessary. Here we show that using optogenetic stimulation of glutamatergic neurons in primary visual cortex (V1) of awake macaque monkeys, pairwise correlations can be modulated independent of firing rate. This technique could be used to test theoretical predictions about the functional impact of correlated variability. We delivered the channelrhodopsin (ChR2) gene to multiple sites in V1 using a lentivirus vector with a CaMKII promoter in two monkeys (*macaca mulatta*). Starting 4 weeks after the injection, single and multi-unit activity were recorded using laminar electrodes and a laser-connected fiber optic cable. Monkeys performed a fixation task and half of the trials were paired with simultaneous optical stimulation (10 pulses at 35 Hz). Only trials with no visual stimulus in which monkeys maintained fixation on a central spot on a computer screen and correctly reported the absence of a visual stimulus were considered. We recorded a total of 1,478 cell pairs across 20 sessions, of which 89% were responsive to the optical stimulation. We computed noise correlations across trials for all pairs of neurons by aligning trials with the onset of the laser pulses. We found that for pairs of neurons in which both cells were responsive to light, the mean firing rates of neurons increased during light presentation, and then returned to baseline almost immediately following a very brief drop 50 ms after laser offset. Interestingly, correlations showed no change during optical stimulation, but dropped significantly below the mean correlation in control trials (without optical stimulation) after laser offset ($P<0.002$, rank sum test, with Bonferroni correction). This significant drop in correlations lasted hundreds of milliseconds, and then returned to baseline. These results have the potential to provide causal methodology to subsequently test the impact of noise correlations on neural coding and behavioral decisions.

Disclosures: **A.R. Andrei:** None. **S. Pojoga:** None. **R. Janz:** None. **V. Dragoi:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.02/M2

Topic: B.10. Network Interactions

Support: IWT Baekeland grant 090279

IWT R&D grants 120511 and 150003

Title: Morphofunctional interrogation of *In vitro* neuronal networks exposes modulators of synaptic connectivity

Authors: *P. VERSTRAELEN[1], M. VERSCHUUREN[1], J. DETREZ[1], R. M. NUYDENS[2], A. EBNETH[2], J.-P. TIMMERMANS[1], W. H. DE VOS[1]
[1]Univ. of Antwerp, Antwerp, Belgium; [2]Janssen Res. & Development, A Div. of Janssen Pharmaceutica NV, Beerse, Belgium

Abstract: Orchestrated activation of genetic programs and spontaneous electrical activity guide the wiring of neuronal networks. Mature networks are characterized by the expression of synaptic markers, synchronized electrical activity and the presence of dendritic spines. While neurodevelopmental disorders are thought to result from defective neuronal network formation, neurodegenerative disorders are characterized by progressive synapse loss. Today, basic knowledge about the underlying mechanisms at the cellular and neuronal network level seems insufficient to develop disease-modifying treatments.
We established a model based on mouse primary and human iPSC-derived neurons that recapitulates features of mature networks [1-3]. An image analysis script was developed in Acapella® (PerkinElmer) to quantify neurite- and synapse density, i.e. colocalizing pre- and postsynaptic puncta. To facilitate dendritic spine analysis in dense cultures, sparse labelling strategies such as AuNP-mediated photoporation were explored. Calcium fluctuations, serving as a proxy for electrical activity, were detected in neurons expressing the GCaMP6f reporter. These readouts were integrated into one classification scheme that accurately predicts culture maturity. Morphofunctional connectivity was analysed at different timepoints after pharmacological or molecular biological perturbation. Using microtubule (MT) stabilizing- and depolymerizing compounds, we found that MT stability is an important mediator of synaptic connectivity [5]. This finding was substantiated by overexpressing Tau forms with different microtubule-stabilizing capacity, and by modulating tubulin acetylation. We also saw that dendritic spine density was decreased by inducing a cellular hypoxia response.
This platform for interrogation of morphofunctional connectivity in neuronal networks represents an attractive model for high-content screening in the context of neurodevelopmental or neurodegenerative disorders.

1. Kuijlaars, J., et al., *Sustained synchronized neuronal network activity in a human astrocyte co-culture system*. Sci Rep, 2016. 6: p. 36529.
2. Detrez, J.R., et al., *Image Informatics Strategies for Deciphering Neuronal Network Connectivity*. Adv Anat Embryol Cell Biol, 2016. 219: p. 123-48.
3. Verstraelen, P., et al., *Pharmacological characterization of cultivated neuronal networks: relevance to synaptogenesis and synaptic connectivity*. Cell Mol Neurobiol, 2014. 34(5): p. 757-76.
5. Verstraelen, P. et al, *Dysregulation of microtubule stability impairs morphofunctional connectivity in cultivated neuronal networks*. Front. Cell. Neurosci (in review).

Disclosures: P. Verstraelen: None. **M. Verschuuren:** None. **J. Detrez:** None. **R.M. Nuydens:** None. **A. Ebneth:** None. **J. Timmermans:** None. **W.H. De Vos:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.03/M3

Topic: B.10. Network Interactions

Support: JSPS KAKENHI 16J02485

Synergy Grant (“BASICS”) by Novonordiskfonden

Title: Two oscillatory modes of introducing cortical plasticity in human motor cortex: a transcranial alternating current stimulation study

Authors: *M. TAKEMI[1,2], J. KESSELHEIM[2], M. G. JØNSSON[2], H. R. SIEBNER[2,3]
[1]Grad. Sch. of Educ., The Univ. of Tokyo, Tokyo, Japan; [2]Danish Res. Ctr. for Magnetic Resonance, Copenhagen Univ. Hosp. Hvidovre, Hvidovre, Denmark; [3]Dept. of Neurol., Copenhagen Univ. Hosp. Bispebjerg, Copenhagen, Denmark

Abstract: Transcranial alternating current stimulation (TACS) is a non-invasive neuromodulatory technique capable of interacting with endogenous brain oscillations. It enables to study the causal role of brain rhythms in motor, sensory and cognitive functions and to induce cortical plasticity in a frequency-specific manner. Recently it has been suggested that neurons are susceptible to low-frequency sinusoidal envelope that modulates the amplitude of a high frequency carrier [1]. Such amplitude-modulated tACS (am-TACS) could avoid contamination of stimulation artifacts into the frequency bands of low-frequency physiological oscillations [2], possibly allowing for online assessment of neural oscillations during TACS. However, it remains to be determined how the physiological effects of am-TACS compare to standard TACS at constant amplitude (con-TACS).

In 20 healthy participants, we applied am-TACS with the amplitude of a 140 Hz carrier being modulated by a sinusoidal envelope at 5, 10 or 20 Hz and con-tACS at 5, 10, 20 and 140 Hz. We assessed the lasting effects of TACS on corticospinal excitability, as indexed by motor evoked potentials elicited by TMS (Experiment 1), and brain oscillations using EEG (Experiment 2). Two 9 cm^2 tACS electrodes were placed over the hand area of left primary motor cortex and the midline of the parietal cortex. Single session consisted of one online block (6 min of 1 mA TACS) and three offline blocks (0−1, 5−6 and 10−11 min after TACS). In Experiment 1, we found online enhancement of the corticospinal excitability by beta (20 Hz) con-TACS and theta-gamma (5 Hz envelope with 140 Hz carrier) am-TACS. However, in Experiment 2 these two interventions induced different online effects for brain oscillations. The beta con-TACS increased beta EEG power on the left sensorimotor and parietal cortices, whereas the theta-gamma am-TACS globally decreased beta EEG power and increased theta EEG power. We also found that, contrary to the previous study [2], EEG data during am-TACS were still contaminated by stimulation artifacts at the frequency of low-frequency envelope.
These results reveal two different modes of inducing corticomotor plasticity by externally applied alternating electric fields. We speculate that beta con-TACS directly resonate with beta oscillations, prevalent in sensorimotor cortex, whereas theta-gamma am-TACS remotely interact with motor cortical activities, resulting in suppression of beta rhythms.
[1] Negahbani et al., SfN abstract 2016; [2] Witkowski et al., Neuroimage 2016

Disclosures: M. Takemi: None. **J. Kesselheim:** None. **M.G. Jønsson:** None. **H.R. Siebner:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Biogen Idec. Denmark A/S. F. Consulting Fees (e.g., advisory boards); Lundbeck A/S.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.04/M4

Topic: B.10. Network Interactions

Support: National Center for Research Resources (P51RR000165)

Research Infrastructure Programs (OD P51OD011132)

Title: Effect of higher alfaxalone anesthesia on brain functional connectivity in rhesus monkeys

Authors: *C. LI[1], D. KEMPF[1], L. HOWELL[1,2], X. ZHANG[1,2]
[2]Div. of Neuropharm. and Neurologic Diseases, Yerkes Natl. Primate, [1]Emory Univ., Atlanta, GA

Abstract: Alfaxalone is suggested to be an optimal anesthetic in preclinical studies and has been increasingly used in non-human primates (NHPs). However, little is known about its impact on neural activity in anesthetized subjects. In the present study, adult rhesus monkeys (n=4, 9-13 years old) were used to examine its impact on brain functional connectivity such as default-mode network (DMN). Alfaxalone was first given as an intramuscular injection (5 mg/kg) followed by intravenous infusion (0.125 or 0.156 mg/kg/min) to anesthetic effect for about 1 hour. Then alfaxalone was stopped and followed with ~0.8% isoflurane to provide continuous anesthesia. Resting state functional MRI (rsfMRI) data were acquired using a multiband EPI sequence (TR/TE=1090 ms/25ms, spatial resolution= $1.5\times1.5\times1.5mm^3$) and started ~15 minutes after animals were moved into the scanner (Siemens 3T Trio with an 8-channel Tx/Rx volume coil). rsfMRI data were processed by FSL and AFNI. The averaged time course of rsfMRI signal in PCC (posterior cingulate cortex) was used for seed-based correlation analysis separately. *Z* transformation was applied to the individual correlation maps to show normalized correlation maps. The averaged *z* values of connectivity between PCC and ACC (anterior cingulated cortex) or DMPFC (dorsal/media prefrontal cortex) were examined for statistical differences. The results demonstrate that high dosage alfaxalone significantly reduces functional connectivity in the dominant DMN such as PCC-ACC ($p<0.05$) and PCC-DMPFC ($p=0.06$) connectivity compared to 0.8% isoflurane administration. Also, high dosage alfaxalone showed a stronger inhibiting effect than low dosage alfaxalone by significantly reducing functional connectivity in PCC-DMPFC ($p=0.05$) but not in PCC-ACC with low dosage alfaxalone. These findings reveal that alfaxalone suppresses neural activity more dramatically than isoflurane anesthesia in experimental animals in a dose-dependent manner, suggesting such dose-dependent effects should be considered when choosing anesthetic protocols to examine neuronal function in neuroimaging.

Disclosures: C. Li: None. **D. Kempf:** None. **L. Howell:** None. **X. Zhang:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.05/M5

Topic: B.10. Network Interactions

Title: Mutations causing alternating hemiplegia of childhood disrupt normal neural network activity patterns

Authors: *K. MCSWEENEY[1], E. HEINZEN[2], M. BOLAND[2], D. GOLDSTEIN[2]
[1]IGM, [2]Inst. for Genomic Med., Columbia Univ., New York, NY

Abstract: Alternating Hemiplegia of Childhood (AHC) is a rare and debilitating neurological disorder affecting one in a million children. Greater than 80% of known cases are caused by de

novo mutations in the *ATP1A3* gene. ATP1A3 is exclusively expressed in neurons and is thought to restore basal ionic concentrations in neurons after periods of fast firing. However, the underlying mechanism of pathogenesis of AHC-causing mutations is still unknown, and there are currently no universally effective treatments. We use a multi-well microelectrode array (MEA) platform to evaluate the spontaneous activity of neuronal cultures possessing AHC-causing mutations. MEAs are powerful tools for detecting spontaneous neuronal activity and are an emerging platform for identifying targeted therapies. We recorded neuronal activity 15 minutes each day to collect spike, burst, and network synchronicity data. Preliminary data gathered from primary cortical cultures from two mouse models of AHC - Myshkin (I810N) and Mashlool (D801N) - indicate aberrant organization of neuronal firing patterns in comparison to primary cultures from wild-type littermates. We hypothesize that aberrant activity is due to an imbalance of inhibitory and excitatory signals in the neuronal cultures. To test this hypothesis, we evaluate the effect of several compounds with known mechanisms of action on the activity of cultured neuronal networks. Any identified compound that corrects the aberrant activity patterns will then be tested *in vivo* in the mouse models. Preliminary data indicate a strong shift in the response of mutant neuronal cultures to ATP, providing evidence for decreased ATPase activity in mutant cultures. These experiments illustrate the utility of the MEA platform in the evaluation of genetic mutations in rare neurological disorders and the response of such cultures to compounds. This platform will be used to identify candidate targeted therapeutics for patients with AHC.

Disclosures: K. McSweeney: None. **E. Heinzen:** None. **M. Boland:** None. **D. Goldstein:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.06/M6

Topic: B.10. Network Interactions

Support: NCBS/TIFR

UGC/ISF No.F 6-18/2014(IC)

Title: Subthreshold divisive normalization controls gain and timing in hippocampus

Authors: *S. MOZA, A. BHATIA, U. S. BHALLA
Natl. Ctr. For Biol. Sci., Bangalore, India

Abstract: We report that tightly balanced feed-forward inhibition performs Subthreshold Divisive Normalization (SDN), and controls the peak timing of post-synaptic potentials (PSPs) in the hippocampal CA3-CA1 circuit. SDN is a novel form of gain control, which expands the

range of inputs that a single neuron can accommodate before it reaches spike threshold. While there are several reports of divisive normalization at the firing rate level in sensory cortices (Olsen, 2008; Carandini, 1997), this is the first report, to our knowledge, of such a mechanism at the subthreshold level.
We used a DLP projector (60Hz refresh rate) to project 16um x 16um spots (10ms pulse) on channelrhodopsin-2 expressing CA3 neurons in acute mouse hippocampal slices. Each stimulus combination produced a PSP, which we read out from a patched CA1 neuron. In order to explore a wide dynamic range of inputs, we presented > 100 combinations of 1, 2, 3, 5, 7, or 9 input spots to each cell. We first presented single spots individually and calculated sums of their PSPs (linear sum). We then presented combinations of spots simultaneously, and measured their response (observed sum).
We compared the linear sum from individual spots to the observed sum of the corresponding spots presented together. It turns out that over a large range of inputs, the responses fit a divisive normalization model better than subtractive or divisive inhibition. Blocking inhibition in the slice abolishes SDN.
To further understand the role of inhibition in the mechanism of SDN, we went on to look at the relationship between excitation and inhibition for every single combination of projected spots. Surprisingly, we discovered that excitation and inhibition were very tightly balanced for every one of those combinations. We also built a conductance model which supports divisive normalization in a tightly balanced feedforward inhibitory network. Our observations are in agreement with theoretical ideas of tight E/I balance (Deneve, 2016; Vogels, 2009), which predict that neurons in a tightly balanced network are efficient temporal coders.
While SDN allows integration of a large range of inputs, at large input amplitudes, SDN results in saturation of PSP peaks. The accompanying saturation implies loss of information of input amplitudes. Interestingly, we find that this information is not lost, but simply transferred from amplitude to time of peak of the PSP, with larger number of inputs leading to earlier time to peak. This is a way by which spike timing can be controlled at millisecond time scales, which is relevant to phenomenon like hippocampal replay of memories (Jadhav, 2011).

Disclosures: **S. Moza:** None. **A. Bhatia:** None. **U.S. Bhalla:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.07/M7

Topic: B.10. Network Interactions

Support: The Hartwell Foundation

Title: Spontaneous neural dynamics and functional connectivity in cortical circuits across timescales

Authors: *R. FERNANDEZ GALAN[1], N. KODAMA[2], T. FENG[3], J. J. ULLETT, 44106[3], S. S. SIVAKUMAR[3]
[1]Dept Neurosci, [2]Dept. of Neurosciences, [3]Case Western Reserve Univ., Cleveland, OH

Abstract: Spontaneous activity in brain circuits provides a means to investigate neuronal interactions and measure their functional connectivity, which is crucial to understand how the brain processes information. We investigated spontaneous neural activity in cortical circuits from the mouse neocortex *in vitro* using high-density, multielectrode arrays (MEA). The arrays consist of 120 electrodes evenly spaced over an octagonal area of 1.2 squared mm, which enables the study of activity patterns with unprecedented temporal and spatial resolutions across the six cortical layers over a few cortical columns. Using spike-sorting algorithms, action potentials from multiple cells were resolved from each electrode in the MEA. This allowed us to investigate the spontaneous activity and interactions of hundreds of neurons for up to 30 min. We found that cortical neurons display very diverse firing patterns across multiple frequencies and timescales: some neurons fired tonically, some transiently, and some in regular bursts. The timescales of bursting ranged from milliseconds to several seconds. To investigate the functional connectivity of cortical circuits across timescales, we first treated the spike trains as count processes, that is, we “binned” the firing of each neuron using bins of increasing duration, from 1 ms to 32 s across 20 timescales evenly spaced in logarithmic units. We then computed the cross-correlations in the spike count across neurons for each timescale. On fast timescales, up to the order of a second, interactions between neurons are dominated by sparse and positive correlations. In contrast, on slower timescales, up to the order of tens of seconds, cortical circuits self-organize into two competing, heterogeneous networks with negatively correlated firing fluctuations. One of these networks co-localizes with layer 2/3; the other with layer 5/6. In terms of network properties, the assortativity of functional connections increases dramatically on slower timescales, and the number of functional connections is maximal on intermediate timescales on the order of 1-2 seconds. Altogether, our results demonstrate that neuronal interactions in cortical circuits are qualitative and quantitatively different on different timescales.

Disclosures: R. Fernandez Galan: None. **N. Kodama:** None. **T. Feng:** None. **J.J. Ullett:** None. **S.S. Sivakumar:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.08/M8

Topic: B.10. Network Interactions

Support: Swiss National Science Foundation

Title: The role of local excitatory networks in the lateral amygdala in emotional memory learning

Authors: *M. ABATIS[1], R. NIU[1], R. PERIN[2], G. GIOBELLINA[1], H. BITO[3], H. MARKRAM[4], R. STOOP[1]
[1]Dept. of Psychiatry, CHUV, Prilly, Switzerland; [2]Brain Mind Institute, EPFL, Lausanne, Switzerland; [3]Univ. Tokyo Grad Sch. Med., Tokyo, Japan; [4]EPFL, Blue Brain Project, Lausanne, Switzerland

Abstract: Introduction: Fear conditioning combines an unconditioned stimulus with a conditioned stimulus (CS) so that the CS alone can subsequently elicit fear-related responses. While the convergence of signals onto single lateral amygdala (LA) neurons has been extensively studied, little is known about how a putative local network could perform memory encoding and recall through pattern completion. Recent findings in network organization of the hippocampal CA3 suggest that sparse networks are capable of pattern completion (Guzman et al., Science, 2016).
Aims: We hypothesized that threat-related signals are re-integrated in the LA through local neuronal assemblies. To address this, we aimed to characterize the LA's network organization, and plasticity-induced changes.
Methods: We used whole-cell patch-clamp recordings to simultaneously access up to 12 neurons at a time. The connectivity of over 563 neurons was assessed by delivering, successively, trains of 8 pulses at 20 Hz and monitoring for induced post-synaptic potentials. Neuronal memory-recruitment was assessed by expressing either a destabilized GFP or Channelrhodopsin-2 (Tamoxifen-inducible Cre-recombinase) under an enhanced Arc promoter, after threat memory recall.
Results: We observed ~2% connectivity biased towards close-proximity neurons. Analysis of the peak excitatory post-synaptic current amplitudes by simple binomial analysis suggested 1-5 release sites, with a quantal size of 10±3 pA and probability of release of ~0.5 ± 0.2 (±SD). Furthermore, these connections were amenable to spike-timing-dependent plasticity which led to a 30% increase in excitatory post-synaptic potential (EPSP) amplitude. Next, we assessed plasticity among neurons participating in the recall of a threat memory trace. Recruited neurons had higher probability to observe an EPSP (0.5±0.04 %) compared to controls (0.4±0.01%; ±s.e.m.). This suggests that either more efficient connections enhance local network excitability and favor memory recruitment or that the observed changes result from memory recruitment. Finally, we investigated the contribution of memory-recruited neurons in pattern completion *in vivo*. We found that both CS and optogenetic activation - but not a neutral tone - could reproduce similar activation patterns.
Conclusions: The LA network follows a "small-world" network organization, with memory-recruited neurons forming stronger connections capable of pattern completion. This suggests either that enhanced local network excitability favors memory recruitment or that neurons are biased to be recruited among efficient network nodes, to be determined by future experiments.

Disclosures: **M. Abatis:** None. **R. Niu:** None. **R. Perin:** None. **G. Giobellina:** None. **H. Bito:** None. **H. Markram:** None. **R. Stoop:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.09/M9

Topic: B.10. Network Interactions

Support: CONACyT Grant 414165

PAPIIT-UNAM Grant IN200415

CONACyT Grant 164536

Title: Changes of intracellular calcium concentrations induced by oxytocin and vasopressin on dorsal horn cells from spinal cord slices: Implications in nociception modulation

Authors: *I. TELLO-GARCÍA[1], G. MARTÍNEZ-LORENZANA[1], A. GONZÁLEZ-HERNÁNDEZ[1], J. PÉREZ-ORTEGA[2], J. BARGAS[2], M. CONDÉS-LARA[1]
[1]Univ. Nacional Autónoma De México, Queretaro, Mexico; [2]Inst. De Fisiología Celular - Univ. Nacional Autónoma De México, México, Mexico

Abstract: Recently, oxytocin (OT) and vasopressin (AVP) have been studied as potential modulators of endogenous analgesia. At spinal level the precise molecular mechanisms of this modulation are still unknown. Some studies suggest that such modulation involves the oxytocin receptor(OTR) while others support the vasopressin receptor (V1aR) hypothesis. In this way, it is unknown how these neuropeptides alter the dynamics of intracellular calcium concentrations, $[Ca^{2+}]_{i,}$ to modulate the transmission of information in the spinal cord cells.
As an initial approach, the aim of this study was to evaluate the effect of OT and AVP on intracellular Ca^{2+}-dependent cellular activity of the dorsal horn of the spinal cord. We used Ca^{2+} video-microscopy analysis on lumbar portion of spinal cord slices of neonatal rats. To identify projection neurons populations, the rats were previously injected with neuronal retrograde tracer (dextran tetramethylrhodamine) in the Gracile nucleus or in the ventral posterolateral thalamic nucleus (VPL). The slices were incubated with Fluo-8AM® and subsequently videos of spontaneous activation on spinal dorsal horn was recorded before (spontaneous or basal activity) and after OT (5 μM) or AVP (5 μM). Data analysis was carried out using algorithms written in LabView® and MatLab® where changes in fluorescence of regions of interest (ROI's) corresponding to cells were measured and normalized to evaluate their change over time. Also, coactivity, clustering and connectivity analysis were performed to estimate the changes in activation of cell populations over time.

Cellular activation of the spinal cord was modified by both OT (5 μM) and AVP (5 μM). OT produces an increase on intracellular Ca^{2+} concentrations mainly in superficial laminae (I-III) of the dorsal horn, promoting the: (i) coactivation of groups of cells, (ii) formation of microcircuits, and (iii) interaction between them. In contrast, AVP induces a decrease on intracellular Ca^{2+}-dependent activation along the dorsal horn laminae (I-VI). In addition, AVP abolishes the coactivation of groups of cells and the interaction between them. The results suggest that both OT and AVP modulate changes in cellular activation dynamics dependent of intracellular Ca^{2+} variations, changes that could be responsible for the antinociceptive functions attributed to these peptides. However, it is necessary to evaluate and identify how the activation dynamics of specific cellular populations changes by the OTR or V1aR activation.

Disclosures: I. Tello-García: None. **G. Martínez-Lorenzana:** None. **A. González-Hernández:** None. **J. Pérez-Ortega:** None. **J. Bargas:** None. **M. Condés-Lara:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.10/M10

Topic: B.10. Network Interactions

Support: 1DP5OD012109

1R03MH105765

1R01MH107426

TL1 TR000141

BlackThorn Therapeutics

Swartz Foundation

Title: Hierarchical heterogeneity of circuit properties across human cortex shapes multiple temporal scales of spontaneous dynamics

Authors: *M. DEMIRTAS[1], M. HELMER[2], J. B. BURT[3], J. JI[3], S. SOTIROPOULOS[5], A. ANTICEVIC[4], J. D. MURRAY[2]
[1]Psychiatry Dept., [2]Dept. of Psychiatry, [4]Psychiatry, [3]Yale Univ., New Haven, CT; [5]Univ. of Oxford, Oxford, United Kingdom

Abstract: A central challenge in systems neuroscience is to understand how the large-scale organization of brain function emerges from structured long-range interactions among local neural circuits. Noninvasive neuroimaging studies, such as fMRI, have shown that human brain

is organized into large-scale networks. Yet neuroimaging cannot directly inform the state of cortical physiology. One approach to bridge this gap is to develop biophysically-based models of large-scale brain circuits that capture key features of neuronal and synaptic dynamics. These models can be integrated with multimodal human neuroimaging constrained by structural connectivity, and optimized to match functional dynamics. Early modeling studies have focused on how structural connectivity shapes patterns of resting-state functional connectivity (rs-FC). However, it remains unclear how local circuit properties, and their heterogeneity across brain areas, contribute to these large-scale dynamical patterns. To investigate these issues, we developed a large-scale cortical model of rs-FC that incorporates heterogeneity of local microcircuit properties along the cortical hierarchy (from sensory to association cortex). We parametrized hierarchical differences of circuit properties using a noninvasive MRI-derived measure of structural heterogeneity. To test the model, we harnessed a state-of-the-art multimodal dataset from the Human Connectome Project (HCP): resting-state BOLD, structural connectivity of inter-areal projections derived from Diffusion-Weighted Imaging, maps of local structural heterogeneity, and resting-state MEG. We found the ability of the model to quantitatively fit empirical BOLD rs-FC patterns is substantially improved with hierarchical specialization of circuit properties, specifically with a gradient of increased local recurrent strength along the hierarchy. Furthermore, hierarchical heterogeneity allows the model to capture empirically observed inter-individual variability in rs-FC patterns. Beyond rs-FC of the BOLD signal, the underlying synaptic activity of the heterogeneous model predicted regional heterogeneity in the spectral properties of higher-frequency neural activity. We found that the topography of spectral variation predicted by the model matches to empirical measurements from MEG. Our findings suggest that heterogeneity of local circuit properties across the cortical hierarchy plays an import role in shaping the large-scale organization of resting-state cortical dynamics.

Disclosures: **M. Demirtas:** None. **M. Helmer:** None. **J.B. Burt:** None. **J. Ji:** None. **S. Sotiropoulos:** None. **A. Anticevic:** F. Consulting Fees (e.g., advisory boards); BlackThorn Therapeutics. **J.D. Murray:** F. Consulting Fees (e.g., advisory boards); BlackThorn Therapeutics.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.11/N1

Topic: B.10. Network Interactions

Support: 1DP5-OD012109

TL1 TR000141

BlackThorn Therapeutics

Title: Hierarchical organization of microcircuit specialization across human cortex captured by myelin topography

Authors: *J. B. BURT[1], M. DEMIRTAS[1], W. J. ECKNER[1], J. WANG[1], N. NAVEJAR[2], L. JI[1], A. BERNACCHIA[3], A. ANTICEVIC[1], J. D. MURRAY[1]
[1]Yale Univ., New Haven, CT; [2]Tulane Univ., New Orleans, LA; [3]Univ. of Cambridge, Cambridge, United Kingdom

Abstract: Hierarchy is a general principle underlying the large-scale organization of primate cortex that describes differences in structural and functional properties across cortical areas. Structural hierarchy is determined by the laminar patterns of interareal projections, as measured by anatomical tracer studies. Functional specialization, including in representational selectivity and temporal dynamics, has been found to correlate with structural hierarchy. Yet the neural circuit basis of areal specialization remains an open question. It remains unclear how cortical microcircuit properties vary with hierarchy, especially in the human, where structural hierarchy cannot be measured through invasive tracer studies.
In this study, we validated a noninvasive neuroimaging correlate of structural hierarchy, and found that it captures the dominant spatial pattern of variation in gene expression across human cortex. One MRI-derived measure of regional heterogeneity in cortex is the myelin map, which reveals intracortical differences in myelin content. We found that in the macaque monkey, the myelin map predicts anatomically-defined hierarchical relationships among cortical areas. This validates the myelin map as a noninvasive neuroimaging-based proxy measure for hierarchy which can be applied to human cortex.
Leveraging the human myelin map, we examined hierarchy-related patterns of gene expression variation across human cortex using the Allen Human Brain Atlas. We observed hierarchical gradients in expression profiles for genes regulating synaptic receptor subunit composition, and hierarchical gradients in genes implicated in neuronal subtype specificity, which we show follow trends present in quantitative anatomical measurements in the macaque. Moreover, human cortical myelin map topography was remarkably similar to the first principal component of gene expression variation, indicating that myelin maps capture the dominant mode of gene expression variation across human cortex. Furthermore, we found that risk genes for neurodegenerative disorders exhibit hierarchically graded expression patterns, highlighting the importance of hierarchy for understanding cortical function.
Our findings suggest that hierarchy is a general principle of large-scale cortical organization in primates that underlies structural and functional specialization of local cortical microcircuits. Cortical hierarchy thereby provides a rich theoretical framework for relating human brain function to circuit physiology.

Disclosures: **J.B. Burt:** None. **M. Demirtas:** None. **W.J. Eckner:** None. **J. Wang:** None. **N. Navejar:** None. **L. Ji:** None. **A. Bernacchia:** None. **A. Anticevic:** F. Consulting Fees (e.g., advisory boards); BlackThorn Therapeutics. **J.D. Murray:** F. Consulting Fees (e.g., advisory boards); BlackThorn Therapeutics.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.12/N2

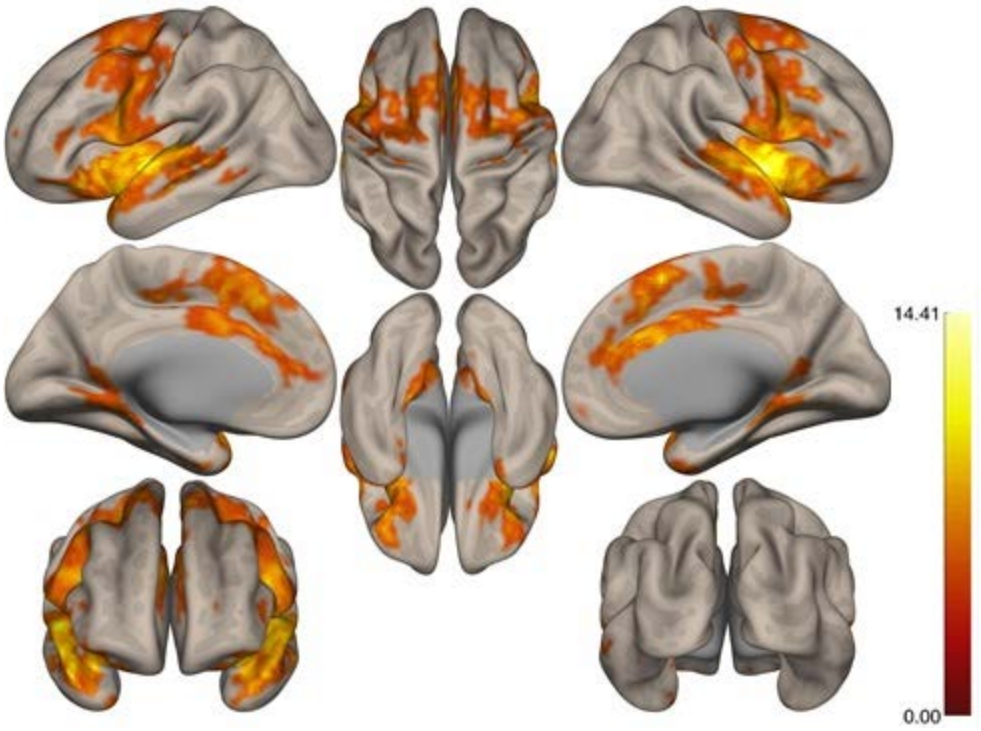
Topic: B.10. Network Interactions

Support: UNAM DGAPA-PAPIIT IN203216

Title: Functional connectivity of the human claustrum

Authors: *F. A. BARRIOS, L. RODRÍGUEZ, S. ALCAUTER
Univ. Nacional Autonoma de Mexico, Queretaro, Mexico

Abstract: The claustrum is a bilateral, small cerebral structure integrated mainly by somas. It is located between the capsula extrema and the putamen's capsula externa. Structural brain connectivity studies show the claustrum is profusely connected to cortical and sub-cortical structures. High resolution tractographyc studies described 4 main groups of neural tracts; anterior projecting to the prefrontal cortex, posterior projecting to the visual cortex, superior to the sensorimotor cortex and lateral to the auditory cortex. Presently, it has not been determined the claustrum's main function, beyond a relay capacity, due its abundant cortical and sub-cortical connectivity. In this work, we present a high resolution functional connectivity study based on 40 resting state studies taken from the Human Connectome Project (HCP, NIH Bleuprint: The Human Connectome Project), with 2x2x2 mm^3 voxel resolution, equivalent to the high resolution tractographyc studies. Functional connectivity was estimated with seed to voxel correlations from anterior and posterior carefully constructed anatomical seeds, placed at the claustrum using Conn (V17, Functional connectivity toolbox, NITRC). Functional connectivity of the claustrum was found ($p<0.05$, p-FDR corrected) with the right and left temporal pole, the right and left pre-central gyri, right and left insular cortex, the anterior cingulate cortex, the superior pre-central gyrus, orbitofrontal cortex, among others. We estimated a general claustrum network shown in the figure, also an anterior posterior segmentation of the functional connectivity of the claustrum was estimated.



Disclosures: **F.A. Barrios:** None. **L. Rodríguez:** None. **S. Alcauter:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.13/N3

Topic: B.10. Network Interactions

Title: Rhythmic activity of astrocytes synchronized with alternating motor output during fictive locomotion

Authors: *I. YAZAWA[1], S. OKAZAKI[2,3], S. YOKOTA[4], K. TAKEDA[5,3], H. MIKAMI[3], I. FUKUSHI[3], H. ONIMARU[6], Y. OKADA[3]
[1]Global Res. Ctr. for Innovative Life Sci., Hoshi Univ. Sch. of Pharm. & Pharmaceut. Sc, Shinagawa, Japan; [2]Waseda Univ., Tokorozawa/Saitama, Japan; [3]Clin. Res. Ctr., Murayama Med. Ctr., Tokyo, Japan; [4]Dept. of Anat. and Morphological Neurosci., Shimane Univ. Sch. of Med., Izumo, Japan; [5]Sch. of Hlth. Sci., Fujita Hlth. Univ., Toyoake, Aichi, Japan; [6]Showa Univ. Sch. of Med., Tokyo, Japan

Abstract: Locomotor pattern generator (LPG) in mammals has remained a ‘black box’ where inputs to the network were manipulated and the outputs interpreted. However, it has been already

known that spinal interneurons are organized into networks that control the activity and output of the motor system. Recently, it has been elucidated that not only neurons but also glial cells, especially astrocytes, play active roles in various functions in the brain and spinal cord, such as rhythm generation of respiration and mastication. We applied electrophysiological and Ca imaging techniques to the isolated lumber cord preparations and investigated whether not only neurons but also astrocytes are actively involved in generation of rhythmic locomotor-like activities in the LPG during fictive locomotion. Rhythmic locomotor-like neural activity was induced by adding 5-HT and NMDA to the superfusate, and activities of cells in the L5 ventral horn were recorded with a Ca imaging system together with rhythmic neural output from the L4/5 ventral root. Recorded cells were classified into neurons and astrocytes by lowering the superfusate potassium concentration in the presence of TTX (Dallwig and Deitmer, 2002). The detected activities in the neurons and astrocytes, and the output activity were analyzed by cross correlation and causality analyses to illuminate the interaction among the activities of the astrocytes, neurons, and locomotion-like neural output. We found that fractions of not only neurons but also astrocytes revealed rhythmic activities in synchrony with locomotor-like neural output. It was suggested that not only neurons but also astrocytes are involved in the formation of the rhythm and pattern of locomotor neural output in the developing lumbar cord.

Disclosures: I. Yazawa: None. **S. Okazaki:** None. **S. Yokota:** None. **K. Takeda:** None. **H. Mikami:** None. **I. Fukushi:** None. **H. Onimaru:** None. **Y. Okada:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.14/N4

Topic: B.10. Network Interactions

Support: SFB Grant 1089

Title: Effect of induced hippocampal knockout of Ryanodine Receptor 2 on CA1 cellular activity

Authors: *M. MITTAG, L. WISCHHOF, F. BERTAN, L. SOSULINA, S. REMY, D. BANO, P. NICOTERA, M. FUHRMANN
German Ctr. of Neurodegenerative Dis. (DZNE), Bonn, Germany

Abstract: Calcium (Ca^{2+}) signaling, mediated by entry of Ca^{2+} through the plasma membrane or release from the endoplasmic reticulum (ER), plays an important role in neuronal function. Elevation of intracellular Ca^{2+} triggers release of neurotransmitters at the synapse, contributes to dendritic spikes, induces activity-dependent changes in gene expression and regulates synaptic plasticity. Although much of the entry of Ca^{2+} into neurons occurs through plasma membrane

channels, the ER is the major dynamic pool for intracellular Ca^{2+}. Release of Ca^{2+} from the ER into the cell is mediated by the family of Ryanodine Receptors (RyR). RyR2, a RyR isoform, is abundant in the hippocampus and considering its role in Ca^{2+} homeostasis this raises questions about its contributions to learning and memory processes. We want to investigate the effect of a local knockout of RyR2 in the hippocampus on cellular activity of principal neurons in CA1 and if knockout results in deficiencies in hippocampus-dependent learning. By stereotactic injection of an AAV-based virus carrying the gene for cre-recombinase into the hippocampus, we induce the specific knockout of RyR2 in a RyR2 flx/flx mouse model. We tested learning in a Novel Object Recognition (NOR) task and a Radial Arm Maze (RAM). The combination of two-photon microscopy, a chronic hippocampal window and virally expressed Ca^{2+}-indicators allows the recording of the activity of large neuronal populations in awake animals. We could observe spatially tuned firing in CA1 cells in RyR KO mice. For the future, we aim to further characterize the firing properties of hippocampal neurons in the RyR KO model. We also want to investigate if there are functional alterations that relate to performance in learning tasks.

Disclosures: **M. Mittag:** None. **L. Wischhof:** None. **F. Bertan:** None. **L. Sosulina:** None. **S. Remy:** None. **D. Bano:** None. **P. Nicotera:** None. **M. Fuhrmann:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.15/N5

Topic: B.10. Network Interactions

Support: R01MH098003

R01NS085200

Title: A chemogenetic-rsfMRI study in awake rats revealed the inactivation of anterior cingulate cortex in the brain functional network

Authors: *W. TU[1,2], Z. MA[1], Y. MA[1], D. CHAU[1], Y. LIU[1], N. ZHANG[1,2]
[1]Pennsylvania State Univ. Main Campus, University Park, PA; [2]The Huck Inst. of the Life Sci., University Park, PA

Abstract: Brain functional network can be described as a collection of nodes (brain regions) connected by edges (functional connectivity). Similar to human studies, connectivity-based parcellation analysis of brain functional network on awake rats exhibited a topological organization with specialization and integration. Cingulate cortex is crucial for mediating emotion, cognition, and motion. It is recognized as a functional hub region of resting-state rats, characterized by its high degree of connectivity to other brain regions. Inhibition of a hub node

has a profound impact on the brain functional network. However, our understanding on the role of anterior cingulate cortex in the whole brain level was mostly limited to anatomical research. The present study combined the designer receptors exclusively activated by designer drugs approach and resting-state functional magnetic resonance imaging (chemogenetic-rsfMRI) to elucidate the impact of anterior cingulate cortex inhibition on the brain functional network in awake animals. We introduced genetically encoded modified muscarinic G-protein-coupled receptors hM4Di into dorsal anterior cingulate cortex (dACC) using adeno-associated viral vector. Clozapine-N-oxide (CNO) was injected intraperitoneally (IP) 30-45min before the rsfMRI scan to drive the expression of hM4Di in dACC. Functional blood-oxygenated-level dependent signal was measured to assess the dynamics on functional connectivity. Using the Chemogenetic-rsfMRI approach, we were able to reveal the functional circuit underlying the dACC-related processes in awake state.

Disclosures: W. Tu: None. **Z. Ma:** None. **Y. Ma:** None. **D. Chau:** None. **Y. Liu:** None. **N. Zhang:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.16/N6

Topic: B.10. Network Interactions

Support: German Research Foundation (DFG) SCHE 1575/1-1 & 3-1

Title: Behavior dependent antagonistic synchronization of beta and low frequency hub neurons in the macaque fronto-parietal grasping network

Authors: *S. SHESHADRI, B. DANN, H. SCHERBERGER
German Primate Ctr., Goettingen, Germany

Abstract: Synchronization in different frequency bands has been identified as an important mechanism for selective communication within and between different cortical areas. Recent studies in the fronto-parietal network have shown that this synchronization is coordinated by a group of hub neurons. However, it is unknown how network coordination changes with respect to behavior. To this end, we trained two macaque monkeys to perform a delayed grasping task with randomly mixed instructed and free-choice trials, in which a handle had to be grasped with one of two possible grip types. Neuronal activity was recorded in parallel from the fronto-parietal grasping network including the ventral premotor cortex (area F5) and the anterior intraparietal area (AIP) with 64 electrodes chronically implanted in each area. Single unit and local field potential (LFP) activity, representing the local population activity, was extracted from all channels. The degree of synchronization between single units and LFP signals was calculated

using pairwise phase consistency (PPC). Importantly, PPC is a spike-rate independent metric that allows the comparison of different behavioral conditions associated with different single-unit firing rates. The large number of simultaneously recorded units and LFPs allowed us to analyze the synchronization network structure by calculating PPC of all single unit and LFP pairs. Beta (18-35Hz) and low frequency (2-8Hz) bands turned out to be the dominant frequency bands in the network. In beta band, almost exclusively a group of single units in AIP were synchronized with larger populations in both areas. In contrast, synchronization in the low frequency range was largely confined to a group of single units in F5. The average synchronization of single units with LFPs was heterogeneously distributed, confirming that the strongly synchronized groups of units in both frequency bands are hubs. Interestingly, there was hardly any overlap between the two groups although there was a strong overlap of local population activity to which they were synchronized. We found that beta hubs were predominantly synchronized to the network during passive periods (fixation and memory), whereas low frequency hubs dominated synchronization during active periods (cue and movement). In contrast, condition-dependent differences were associated with changes in both beta and low frequency network connectivity. These findings suggest that behavior-relevant changes in the cortical motor network are coordinated by distinct groups of hub units selectively synchronizing network activity in the beta and low frequency range.

Disclosures: **S. Sheshadri:** None. **B. Dann:** None. **H. Scherberger:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.17/N7

Topic: B.10. Network Interactions

Title: Steady-state visual stimulation frequency modulates functional networks

Authors: *Y. CHAI, D. A. HANDWERKER, J. GONZALEZ-CASTILLO, P. A. BANDETTINI
Section on Functional Imaging Methods, NIMH, NIH, Bethesda, MD

Abstract: Background: Studies of the primary visual cortex tend to use stimuli with less than a 20 Hz flickering rate and focus on the occipital cortex. It’s known that steady-state visual evoked potentials (SSVEP) are widely distributed over not only occipital cortex but also multiple functionally distinct brain areas. Additionally, Iaccarino & Singer et al. (2016) showed physiological changes accompanying 40 Hz visual stimuli. We hypothesized that steady-state visual stimulation will drive the large scale functional network changes across the whole brain, and that these changes will dependent on the stimulation frequency. To test these hypotheses, we measured fMRI in humans while applying steady-state visual stimulations at multiple

frequencies, and examine changes in functional networks; as well as their relationship to visual activation. **Methods:** Twelve subjects were scanned on a Siemens 3T system. Steady-state visual stimulation at 4 different frequencies (1 Hz, 10 Hz, 20 Hz and 40 Hz) was applied through reversing the pure black and white screens. Data were collected with continuous stimulation for 386 sec (and a control run with no stimulation) and with a block-designed stimulation (10 sec ON, 20.6 sec OFF). Simultaneous multi-slice (SMS) EPI was used with TR/TE = 1700/28 ms, matrix 90×90, 2.5 mm isotropic voxels, 52 slices, SMS factor = 2. After standard fMRI preprocessing, graph-based modularity analysis was employed to identify different network modules during steady-state visual stimulations, and further investigated the modulation of intra and inter network interactions at different stimulation frequencies. **Results:** At the low stimulation frequency (1 Hz), network strength and efficiency (both local and global) inside visual module is significantly higher in comparison to other higher stimulation frequencies. As the stimulation frequency increased, the network strength and efficiency decreased within the visual module while increased within salience network and sensorimotor modules. The thalamus showed the largest correlation with visual module at 10 Hz. Subjects with stronger correlations between the thalamus and visual module at 40 Hz, also have larger amplitude responses of visual activation to 40Hz block stimulation.

Disclosures: Y. Chai: None. **D.A. Handwerker:** None. **J. Gonzalez-Castillo:** None. **P.A. Bandettini:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.18/N8

Topic: B.10. Network Interactions

Support: MOST grant 105-2633-B-007-001

MOST grant 105-2311-B-007-012-MY3

Title: The neural circuit mechanism of spatial orientation in *Drosophila* -- connectomic analysis and modeling

Authors: *C. LO[1], T.-S. SU, 300[2], W.-J. LEE[2], Y.-C. HUANG[3], C.-T. WANG[3]
[1]Natl. Tsing Hua Univ., Hsinchu, Taiwan; [2]Inst. of Systems Neurosci., [3]Natl. Tsing Hua Univ., Hsinchu city, Taiwan

Abstract: The ability of maintaining spatial orientation is crucial for an animal to perform goal-directed movements. Recent *Drosophila* studies have revealed the critical role of the ellipsoid body (EB) in tracking spatial orientation, but the precise circuitry and underlying mechanisms

remain unclear. We analyzed connectomic data of *Drosophila* central complex and discovered that the circuit connecting EB and the protocerebral bridge (PB) form symmetric and asymmetric rings. The asymmetric rings can be further divided into two sub-rings, one with counterclockwise and the other with clockwise patterns. Interestingly, the circuits involving the sub-rings are delicately organized in a way that each sub-ring can be individually activated by a pair of neurons that innervate PB. We further constructed a spiking neural circuit model based on the circuits reconstructed from the connectomic data. We demonstrated that the symmetric ring is capable of sustaining persistent neural activity that encodes spatial orientation, while the asymmetric rings perform angular path integration and update orientation when the body rotates in the dark. The model reproduces several key features of EB activity and makes experimentally testable predictions, providing new insight into how spatial orientation is maintained and tracked at the cellular level.

Disclosures: C. Lo: None. **T. Su:** None. **W. Lee:** None. **Y. Huang:** None. **C. Wang:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.19/N9

Topic: B.10. Network Interactions

Title: Preliminary investigation of pain-related changes in cerebral blood volume in patients with neuropathic pain after scrambler therapy

Authors: *S. JOO, C. SEO
Hangang Sacred Heart Hosp., Seoul, Korea, Republic of

Abstract: OBJECTIVE:
Pain Scrambler therapy is a patient-specific electrocutaneous nerve stimulation device. Post burn neuropathic pain is often difficult to treat effectively. The neuropathic pain may result from injury to sensory nerves that hampers conductance of neuronal messages along the large A and small C afferent fibers to the spinal cord. We utilized MRI to measure CBV in patients who treated by Scrambler therapy following burn injury to investigate changes in the pain network associated with neuropathic pain.
INTERVENTIONS
The intensity of neuropathic pain was measured using the Visual Analogue Scale (VAS). 8 subjects were recruited to participate in this study. The subjects complained of severe neuropathic pain that was rated at least 5 on the visual analogue scale (VAS), despite treatments with medications and physical modalities. Each Scrambler Therapy with the MC-5A Pain Scrambler Therapy® technology device was performed for 45min daily (Monday through Friday) for 10 consecutive days. The stimulus was increased to the maximum intensity bearable by the

individual patient without causing any additional pain or discomfort. Voxel-wise comparisons of relative CBV maps were made between before Scrambler therapy and after 10 sessions Scrambler therapy over the entire brain volume. The relationship between individual participant CBV (measured in voxels) and VAS score was also examined.

RESULTS

Compared to CBV before Scrambler therapy, the patients had a decrease CBV on primary somatosensory cortex, secondary somatosensory cortex, insular cortex(IC) and anterior cingulate cortex [ACC]) after 10 sessions Scrambler therapy.

CONCLUSIONS

We observed changed CBV in regions associated with cerebral pain network of patients who had undergone burn injury. This study suggests that CBV changes were related to neuroplasticity associated with neuropathic pain after Scrambler therapy.

KEYWORDS:

Blood volume; Brain; Magnetic resonance imaging; Neuropathic pain, Scrambler therapy

Disclosures: S. Joo: None. **C. Seo:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.20/N10

Topic: B.10. Network Interactions

Title: Parvalbumin interneurons control spike output in deep layer principal neurons in the *In vitro* mouse perirhinal - entorhinal cortex

Authors: *N. L. CAPPAERT[1], J. G. WILLEMS[2], W. J. WADMAN[3]

[1]Univ. Amsterdam, Amsterdam, Netherlands; [2]Swammerdam Inst. for Life Sci., Univ. of Amsterdam, Amsterdam, Netherlands; [3]Univ. Amsterdam, Amsterdam, Netherlands

Abstract: The perirhinal cortex (PER) and the lateral entorhinal cortex (LEC) link the neocortex with the hippocampus and are involved in cognitive functions like learning and memory and object recognition. Even though anatomical connections from the neocortex to the hippocampus exist, the information transfer through the PER/LEC network occurs with a low probability. It is hypothesized that this gating is most likely controlled by inhibitory interneurons and this study investigated the role of parvalbumin positive (PV) interneurons located in the deep layers of the PER and LEC.

The response to stimulation of the adjoining superficial agranular insular cortex (AiP) was measured in PV interneurons and principal neurons in the deep layer PER/LEC network in acute horizontal mouse brain slices in whole cell patch recordings. Action potential firing was rarely observed in principal neurons in response to AiP stimulation (6% of the cases) confirming the

hypothesis that information transfer through this network occurs with a low probability. Linear decomposition of the evoked synaptic currents was performed to separate the excitatory from the inhibitory conductance. AiP stimulation resulted in a small excitatory conductance and a substantially larger inhibitory conductance in principal neurons. Simultaneous recordings of a principal neuron and an adjacent PV interneuron were performed to address the origin of this large inhibitory conductance evoked in principal neurons. The same AiP stimulus evoked a larger synaptic input with a shorter latency in PV interneurons compared to the principal neurons. Moreover, the PV interneurons were capable of firing action potentials in response to stimulation.
The aggregated results demonstrated that the inhibitory conductance observed in the principal neurons could well be the result of a response to the spikes in the population of PV interneurons. In conclusion, the excitatory input from the AiP onto deep layer PER/LEC principal neurons is, in these experimental conditions, under strong control of PV interneuron inhibition. Our results indicate an essential gating role for PER/LEC PV interneurons in the signal transfer from the AiP through the PER/LEC network.

Disclosures: **N.L. Cappaert:** None. **J.G. Willems:** None. **W.J. Wadman:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.21/N11

Topic: B.10. Network Interactions

Support: NSERC CREATE

NSERC Discovery Grant #40352

Title: Slow-wave cortical activity within local circuits during REM-like state revealed by fast Voltage-Sensitive Dye imaging

Authors: *M. NAZARIAHANGARKOLAEE, J. KARIMI, M. TATSUNO, M. H. MOHAJERANI
CCBN, Univ. of Lethbridge, Lethbridge, AB, Canada

Abstract: Sleep in mammals consists of two basic stages: non-rapid eye movement (NREM) and rapid eye movement (REM) sleep. NREM sleep can be distinguished by slow and high-amplitude cortical EEG signals while REM sleep is characterized by “desynchronized” low-amplitude fast cortical rhythms. While until recently it has been widely believed that cortical activity during REM sleep is globally desynchronized, using sparse electrophysiological recording a recent study (Funk et al., 2016) showed local slow waves in primary cortical areas

during REM sleep. However, the electrophysiological technique has been unable to resolve the regional structure and dynamics of these activities due to relatively sparse sampling. Here, we investigated local changes in neuronal activity during REM state using mesoscale imaging. Urethane anaesthesia was used as a model of sleep as it induces spontaneous alternation of brain state between REM- and NREM-like in rodents in which cortical and hippocampal activities resemble to those of natural sleep (Clemet et al., 2008). The wide-field VSD imaging from neocortex was combined with LFP recording from ipsilateral hippocampus and motor/somatosensory cortex with a unilateral preparation in anesthetized mice. Generally, urethane anesthesia induced state alternation in VSD signal which was concurrent with LFP state changes. However, in REM-like state slow cortical activity were not globally reduced; while midline and posterior areas typically showed desynchronized patterns, anterior and lateral regions showed synchronized activity. Our data also revealed that synchronized/desynchronized regions during individual REM-like episodes can be dynamic. Our results suggest that brain state alternation under urethane anesthesia could change the cortical ensembles activity reported by VSD imaging and provide evidences for the locality of cortical desynchronization during REM-like state.

Disclosures: M. Nazariahangarkolaee: None. **J. Karimi:** None. **M. Tatsuno:** None. **M.H. Mohajerani:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.22/N12

Topic: B.10. Network Interactions

Support: NIH Brain Initiative Grant R01 EB022903 01

Title: Cortical ensembles based on dendritic plateau generation in the prefrontal cortex

Authors: *S. ANGULO[1], J. W. GRAHAM[1], P. GAO[2], S. DURA-BERNAL[1], S. A. NEYMOTIN[3], S. D. ANTIC[2], W. W. LYTTON[1]
[1]Pharmacol. and Physiol., SUNY Downstate Med. Ctr., Brooklyn, NY; [2]Inst. for Syst. Genomics, Univ. Connecticut Hlth. Ctr., Farmington, CT; [3]Dept. of Neurosci., Brown Univ., Providence, RI

Abstract: Prefrontal cortex (PFC) performs executive functions, which require the selection of relevant information from inputs from multiple brain areas. At the cellular level, cortical pyramidal neurons in Layer 5 of the PFC can produce dendritic plateaus, sustained depolarizations with AMPAr and NMDAr activation in basal dendrites. Our embedded-ensemble encoding theory (EEE) hypothesizes that dendritic plateaus put individual cells in a Standby state. Cells in this state have reduced threshold to firing and can more readily produce

synchronized firing in response to afferent synaptic activity. Cells that are firing together are in the Embedded state, in which ensemble neurons exhibit synchronized firing within the column or across columns. Dendritic plateaus have been well characterized at the cellular level, but their implications at the network level remains unknown. We hypothesize that the generation of dendritic plateaus in the pyramidal neurons will be relevant for the formation of neural ensembles. For this, we modeled and simulated a PFC columnar network with a connectivity that leads to dendritic plateaus, and studied the effects of subpopulations of neurons in Standby or Embedded states. We used the NetPyNE package, which extends the NEURON simulator to facilitate building and parallel optimization of neuronal networks. We built a network simulating the architecture of PFC in rats that includes layers 2/3, 5, and 6. A simplified excitatory pyramidal neuron model was included in L2/3 and L6. An excitatory pyramidal neuron in L5 was optimized to simulate experimental findings that generate dendritic plateaus. Interneurons were added to the model to every layer (parvalbumin, somatostatin, and VIP positive interneurons) with proportions and connectivity according to previous reports. Local excitatory connectivity included mainly AMPA receptors. Long-range input connectivity was designed to target the basal dendrites of L5 pyramidal neurons. Long-range connectivity included AMPA and NMDA receptors with different synaptic weights to obtain dendritic plateaus. L5 pyramidal neurons receiving long-range inputs were able to induce dendritic plateaus and a high frequency rate in comparison with other network subpopulations. The generation of dendritic plateaus induced standby and embedded ensembles in our PFC network model and may be essential for the processing of multimodal cortical information.

Disclosures: **S. Angulo:** None. **J.W. Graham:** None. **P. Gao:** None. **S. Dura-Bernal:** None. **S.A. Neymotin:** None. **S.D. Antic:** None. **W.W. Lytton:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.23/O1

Topic: B.10. Network Interactions

Support: NSF IOS Grant 1354932

Title: Functional and effective connectivity in a motor control center

Authors: *R. FOLLMANN, C. J. GOLDSMITH, W. STEIN
Sch. of Biol. Sci., Illinois State Univ., Normal, IL

Abstract: Nervous systems continuously receive sensory inputs from various modalities that are processed in many neural centers, including premotor circuits that are involved in controlling motor output and action selection. However, little is known about how premotor networks

process multiple individual and concurrent sensory information. Using the crustacean stomatogastric nervous system, we investigate the dynamic restructuring of a premotor circuit under distinct modulatory and sensory conditions, exploring the functional and effective connectivities of the premotor neurons network (Bullmore and Sporns, 2009; Seth et al., J Neurosc, 2015). The stomatogastric motor circuits are under continuous modulatory control by descending projection neurons located in a premotor region that receives various sensory inputs (Stein, J Comp Physiol A, 2009). We have previously characterized the 3D structure of this premotor region, the location of descending neurons, and their contribution to motor pattern selection in different sensory and modulatory conditions (Follmann et al., J Comp Neurology, 2016; Goldsmith et al, Neuroscience Meeting, 2017).
Here, we stimulated chemosensory and mechanosensory pathways known to produce distinct motor outputs (Hedrich et al., J Neurophysiol, 2009), and recorded the neuronal population in this premotor region at individual neuron resolution using voltage-sensitive dye imaging. Experimental data processing consisted of a drift removal function in combination with a nonlinear energy function, and a spike threshold based on the variance of the signal. To assess sensory responses, we first characterized the functional connectivity between neurons using pairwise correlations of their activity responses. The majority of neurons responded to both sensory modalities, with the mechanosensory input consistently eliciting stronger responses. Our analysis indicate that distinct dynamic network structures exist to process the two sensory modalities. For concurrent inputs, a new functional network emerged, distinct from both individual networks, indicating a shared premotor network that is dynamically reconfigured according to changes in sensory conditions. We are currently investigating such dynamic reconfigurations using Granger causality measure (Granger, Econometria, 1969) to infer directed causal interactions between the premotor neurons. The resulting effective connectivity of the premotor neurons may reveal strategic directional influences among the premotor neurons necessary for the functional reconfiguration of the network.

Disclosures: **R. Follmann:** None. **C.J. Goldsmith:** None. **W. Stein:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.24/O2

Topic: B.10. Network Interactions

Support: ARL # W911NF-10-2-0022

Title: Patterns of coherence and incoherence driven by structural connectivity in human connectome

Authors: *K. BANSAL[1], G. LIEBERMAN[2], T. D. VERSTYNEN[3], J. M. VETTEL[2,4,5], S. F. MULDOON[1]
[1]Univ. at Buffalo, SUNY, Buffalo, NY; [2]US Army Res. Lab., Aberdeen Proving Ground, MD; [3]Carnegie Mellon Univ., Pittsburgh, PA; [4]Univ. of Pennsylvania, Philadelphia, PA; [5]Univ. of California, Santa Barbara, CA

Abstract: Dynamic patterns of coordinated activity in human brain networks capture and predict human behavior. Fundamentally, this activity originates as interacting neural populations form domains of coherence and incoherence, a phenomenon referred to as a 'chimera state' in a complex systems framework. The appearance of chimera states is due to an interplay between the characteristic dynamics of the interacting network elements, topology, and coupling functions, but the exact conditions that give rise to these states remain unknown in complex environments. Here, we analyze the emergence of chimera states using brain data across a cohort of thirty individuals. Using diffusion weighted imaging, anatomical white matter connectivity matrices are derived for each individual and used in a data-driven computational model of individual brain dynamics. We observe characteristic differences in chimera patterns across individuals, suggesting that the model serves as a valuable tool to understand between-subject variability. We identify that default mode and subcortical sub-networks of the brain show patterns of chimeras that are similar across subjects. This could indicate a limited role of these regions in driving individual variability.

Disclosures: K. Bansal: None. **G. Lieberman:** None. **T.D. Verstynen:** None. **J.M. Vettel:** None. **S.F. Muldoon:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.25/O3

Topic: B.10. Network Interactions

Support: NIMH Fellowship

Title: Hallmarks of criticality under subsampling

Authors: *Y. KARIMIPANAH, D. PLENZ
NIMH, NIH, Bethesda, MD

Abstract: In the cortex, numerous experimental studies have demonstrated that ongoing or evoked activity in superficial layers of neocortex is scale-invariant in space and time. Such scale-invariance has been hypothesized to be a signature of a critical state; a state at the transition between two phases of short-lasting and long-lasting activity, which conveys several advantages

in information processing. All experimental evidence collected so far, however, suffers from the problem of subsampling, which in simulations has been clearly shown to degrade numerous measures of criticality such as power law distributions in sizes and durations or branching parameter estimates. Given these model prediction of subsampling, experimental identifications of true power law scaling should be rare, which is in contrast to what has been reported. Here we address this apparent contradiction between predicted effects of subsampling from theory and simulations and the established power law scaling in experimental reports. Using a minimal computational model of binary probabilistic neurons with sparse random connectivity, we show that adjusting the temporal resolution of the analysis to that of the average network inter-event interval not only retrieves power law distributions for even a small number of sampled neurons, but it also reveals the scaling relation between their exponents if network dynamics is truly critical. This adjustment, which is commonly applied to experimental findings, was further studied analytically and we discuss potential constraints on its applicability for conventional avalanche analysis techniques. Furthermore, we show, both analytically and numerically, that the compelling measure of 'critical slowing down' does not require a particular temporal resolution, yet is also robust to subsampling effects. Our results show that certain signatures of criticality can be reliably retrieved in critical networks even under subsampling conditions.

Disclosures: **Y. Karimipanah:** None. **D. Plenz:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.26/O4

Topic: B.10. Network Interactions

Title: A brain imaging study of functional MRI using ASSR in bipolar disorder

Authors: *K. TAKAO
Natl. Hosp. Organization Hizen Psychiatric C, Kanzaki-Gun, Japan

Abstract: Introduction: The auditory steady state response (ASSR) is one of the indexes of the neural network function of the hearing processing. There are reported that much abnormalities of the gamma band activity in schizophrenia patients. In the functional MRI (fMRI) using ASSR task, there is reported that the 80-Hz stimulation evoked larger blood oxygenation level dependent (BOLD) responses, compared with the other stimulation frequencies in acute phase schizophrenia. However, there is no fMRI using ASSR task study in mood disorders.Objective: In this study, we examined the reaction of the ASSR task in bipolar disorder in using fMRI.Methods: 20 patients of bipolar disorders and 20 normal controls participated in this study. We presented auditory stimulation using a 4-min-block-design paradigm with 8 blocks of 15 seconds of rest and 15 seconds of stimulation including 15click trains. The stimuli were 1-ms

clicks, presented binaurally as trains of clicks for each frequency (29, 30, 40, and 80-Hz). We measured BOLD signals, using a 1.5-T MRI made in Philips Company. Image processing and statistical analyses were performed using the statistical parameter mapping software SPM12 within MATLAB R2015a.Results: We conducted a group analysis between 20 patients of bipolar disorder and 20 normal controls. There is an interaction effect between BOLD responses of patients and healthy people with 40-Hz and 80-Hz stimuli in left FO frontal opeculum (p=0.000001, uncorrected). There is an interaction effect at 40-Hz and 80-Hz stimuli in Post-Hoc analysis (p=0.00004, uncorrected).Conclusion: These results suggested a left frontal lobe dysfunction in the bipolar disorders patients.

Disclosures: K. Takao: None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.27/O5

Topic: B.10. Network Interactions

Support: NIH T32GM007308

Leon Levy Foundation

Alfred P. Sloan Foundation

American Epilepsy Foundation

Title: The role of parvalbumin- and cholecystokinin expressing interneurons in CA3 pattern completion vs. separation

Authors: *S. K. RASHID[1], M. A. DUFOUR[2], R. ZEMLA[2], J. BASU[1]
[1]Neurosci., [2]Neurosci. Inst., New York Univ. Sch. of Med., New York, NY

Abstract: The hippocampus has long been implicated as a center for encoding and recalling episodic memory related to the knowledge of people, places, objects, and events. Pyramidal cells in area CA3 of the hippocampus are essential for retrieval of contextual fear memories and spatial reference memory. These neurons have been shown to be involved in both pattern completion and pattern separation, two important processes for accurate memory recall. Pattern completion allows for an accurate generalization from partial input (i.e. recognizing a friend from behind) and pattern separation allows for distinguishing overlapping inputs as different (i.e. recalling two similar experiences as different events). CA3 can shift between pattern completion and separation based on the overlap between two comparative inputs, but the cellular and circuit mechanisms controlling these two opposing functions have not yet been elucidated. Activity

dynamics in the highly recurrent CA3 auto-associative network must allow context dependent maintenance of stable and distinct place cell sequences while retaining the flexibility to remap in response to changing environmental demands through plasticity mechanisms. Parvalbumin- and cholecystokinin expressing interneurons drive strong synchronous and asynchronous inhibition respectively upon CA3 pyramidal neuron somata to modulate the CA3 output across different time scales and frequency regimes. To understand how local inhibition in CA3 is recruited, we are using two-photon imaging to characterize behaviorally triggered Ca^{2+} events in these two populations in vivo during a head fixed spatial navigation task where mice are exposed to similar and dissimilar multisensory contexts. Using a loss of function approach we are testing whether PV and CCK interneurons contribute differentially to the stability and plasticity of spatially tuned sequences in CA3. We are testing the hypothesis that modulation of GABAergic input from these interneuron populations plays a potential role in regulating CA3 input-output transformation to mediate the switch from pattern completion to pattern separation functions.

Disclosures: **S.K. Rashid:** None. **M.A. Dufour:** None. **R. Zemla:** None. **J. Basu:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.28/O6

Topic: B.10. Network Interactions

Title: Human iPSC-derived neuronal cultures form synchronously bursting cultures and display a seizurogenic response to excitatory pharmacology

Authors: K. MANGAN, *B. D. ANSON, L. HARMS, E. ENGHOFER, C. CARLSON, C. KANNEMEIER
Ceullar Dynamics, Madison, WI

Abstract: The mammalian brain requires a proper balance between excitation and inhibition (E/I ratio). Imbalances in the E/I ratio are associated with numerous neurological abnormalities; increased E/I ratios result in higher excitability, prolonged neocortical circuit activity, stimulus hypersensitivity, cognitive impairments, and even epilepsy while decreased E/I ratios are linked to stronger inhibitory drive, impaired social interactions, autistic behaviors, and mental retardation. During neuronal development the E/I ratio changes and evolves, with excitation decreasing and inhibition increasing, and any deviations to this natural process may give rise to neurological disorders.
A major challenge in neuroscience research and the identification of new drugs is access to clinically-relevant cell models. However, induced pluripotent stem cell (iPSC) technology enables access to previously unattainable human neuronal cell types. Using this technology, we have generated iPSC-derived frontal cortical neurons of an appropriate, brain-similar E/I ratio

(~80% excitation), that develop and display network-level, synapse-coupled, coordinated neuronal activity *in vitro*. Using multi-electrode arrays (MEA) to measure the electrophysiological activity, we are able to capture periodical and synchronized bursting patterns and analyze these data via numerous parameters (including Poisson and ISI statistics). These neuronal cultures contain excitatory synapses (as shown by HOMER1 and synapsin co-staining) that modulate the synaptically-driven and synchronous bursting behaviors observed on the MEA. Importantly, these signals can be blocked by AP5 and DNQX.
To better understand how excitatory pharmacology alters network-level neuronal interactions, we investigated alterations in the regular bursting properties of glutamatergic neurons after addition of (0.003 to 300 uM dose response) various excitatory agents (e.g. bicuculline, chlorpromazine, pentylenetetrazol, amoxapine, 4-aminopyridine, and glutamic acid), common anti-epileptic drugs (e.g. ganaxalone and valproate), as well as a negative (acetaminophen) and vehicle controls (e.g. ethanol and DMSO). Activity metrics displaying dose-dependent responses with pharmacology include: 'single-channel' Poisson bursts (rate, intensity and duration), 'network-level' ISI bursts (rate, intensity and duration), and synchrony measures. The presented data illustrates how human iPSC-technology can be leveraged to create an unprecedented investigatory space for understanding the intricacies of how excitatory synapses control network-connected populations of human neurons.

Disclosures: **K. Mangan:** A. Employment/Salary (full or part-time):; Cellular Dynamics International. **B.D. Anson:** A. Employment/Salary (full or part-time):; Cellular Dynamics International. **L. Harms:** A. Employment/Salary (full or part-time):; Cellular Dynamics International. **E. Enghofer:** A. Employment/Salary (full or part-time):; Cellular Dynamics International. **C. Carlson:** A. Employment/Salary (full or part-time):; Cellular Dynamics International. **C. Kannemeier:** A. Employment/Salary (full or part-time):; Cellular Dynamics International.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.29/O7

Topic: B.10. Network Interactions

Support: ERA-NET EuroTransBio9 In-HEALTH

BMBF #031B0010B

Title: Cerebrospinal fluid from Dementia With Lewy body patients alters neuronal network function *In vitro*

Authors: *S. THEISS[1], H. KOCH[2], G. ELSEN[2], C. NITURAD[2], W. MAETZLER[3], C. DEUSCHLE[3], H. LERCHE[2], M. DIHNÉ[2]
[1]Inst. of Clin. Neurosci., Univ. of Duesseldorf, Duesseldorf, Germany; [2]Ctr. for Neurol. and Hertie Inst. for Clin. Brain Research, Dept. of Epileptology, [3]Ctr. for Neurol. and Hertie Inst. for Clin. Brain Research, Dept. of Neurodegenerati, Univ. of Tübingen, Tübingen, Germany

Abstract: Neuroactive substances associated with central nervous system (CNS) disease can be transmitted throughout the brain via the cerebrospinal fluid (CSF) compartment (*volume transmission*). CSF samples from patients suffering from traumatic brain injury, anti-NMDA-receptor encephalopathy, mild cognitive impairment and Alzheimer's disease have been shown to induce electrophysiological activity changes when applied to mature networks of rodent neurons cultured on microelectrode arrays (MEAs) equipped with 60 Ti/TiN electrodes (30 µm diameter, 200 µm spacing; Multichannel Systems). In order to establish a neuronal *in vitro* model for functional CSF testing, we exposed hippocampal neuronal networks from E17 mice cultured on MEAs to CSF samples from (1) healthy control subjects, (2) Parkinson's disease (PD) patients without signs of dementia and (3) dementia with Lewy bodies (DLB) patients with fluctuating consciousness. CSF samples were collected by routine diagnostic lumbar puncture, and each group comprised ten individual patients. All participants had normal levels in CSF cell count, albumin and IgG index indicating absence of inflammatory CNS disease and blood-brain-barrier disturbance. DLB patients showed significantly lower CSF levels of $Abeta_{1-42}$ monomers compared to controls and PD patients, while h-tau and p-tau levels associated with neurodegeneration were not significantly different. Three weeks after plating, hippocampal networks on MEAs were functionally mature and exhibited spontaneous neuronal activity (spikes) in synchronous network bursts across all 60 recording electrodes. In each 25-minute experiment with these 21 DIV networks, neuronal network activity was recorded on MEAs first in culture media (5 min), then in artificial CSF (aCSF, baseline, 10 min) and finally human CSF (either control subject, PD or DLB patient; 10 min, adjusted to pH 7.4). Under aCSF, networks showed 2,786±396 spikes/min (mean ± SEM, n=10 CSF samples) in 17±3 network bursts/min. Under human control CSF, activity increased to 5705±868 spikes in 23±4 bursts/min (p=0.014, Mann-Whitney test). While applying CSF from PD patients increased spike activity 2.6-fold from aCSF levels—even more than control CSF (2.1-fold), CSF from DLB patients left aCSF activity patterns rather unchanged. We speculate that this statistically significant difference in network responses to PD and DLB patient CSF may be due to additional functional disease related factors in DLB beyond slow neurodegenerative processes. Neuronal network activity on MEAs may be a useful *in vitro* model for the study of pathological candidate factors within human CSF.

Disclosures: S. Theiss: None. **H. Koch:** None. **G. Elsen:** None. **C. Niturad:** None. **W. Maetzler:** None. **C. Deuschle:** None. **H. Lerche:** None. **M. Dihné:** None.

Poster

381. Network Interactions: Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 381.30/O8

Topic: B.10. Network Interactions

Support: Dr. Miriam and Sheldon G. Adelson Medical Research Foundation

Title: Amacrine cells influence the regenerative capacity of adult retinal ganglion cells

Authors: *Q. FENG[1], Y. LI[1], S. PETERSON[1], H.-Y. GILBERT[1], A. BURGESS[2], L. I. BENOWITZ[1]
[1]Boston Children's Hospital/Harvard Med. Sch., Boston, MA; [2]MIT, Boston, MA

Abstract: Mature retinal ganglion cells (RGCs) fail to regenerate their axons after optic nerve crush (ONC) injury due to intrinsic as well as extrinsic suppressors of growth. In vitro data indicate that during development, RGCs lose their intrinsic ability for robust axon growth and regeneration as a result of contact by amacrine cells (ACs), the inhibitory interneurons of the retina. However, it is not known whether ACs continue to contribute to the poor regenerative ability of mature RGCs. To investigate this question, we used a Cre-inducible diphtheria toxin receptor (DTR) approach to express the DTR in ACs, which can then be ablated upon intravitreal administration of diphtheria toxin. After confirming ≥ 90% loss of ACs in the retina, we investigated axon regeneration in the optic nerve. AC ablation nearly eliminated the inner plexiform layer of the retina, in which RGC dendrites receive synaptic input from ACs and bipolar cells, the excitatory interneurons, but surprisingly did not alter the survival of either RGCs or bipolar cells. More importantly, AC ablation enabled RGCs to regenerate injured axons well beyond the site of nerve damage even in the absence of additional treatment. AC ablation significantly increased phosphorylation of ribosomal protein S6 kinase substrate S6 in RGCs, indicating that the presence of ACs normally suppresses S6 kinase activation. Deleting the gene for phosphatase and tensin homolog (pten) in RGCs, another pro-regenerative treatment partially through increasing the activity of S6 kinase, did not synergistically enhance the effect with ACs ablation, whereas other treatments (e.g. zymosan) were complementary. Our results suggest that amacrine cells may exert a suppressive effect on RGC's growth capacity in the adult retina, in part by suppressing S6 kinase activation.

Disclosures: Q. Feng: None. **Y. Li:** None. **S. Peterson:** None. **H. Gilbert:** None. **A. Burgess:** None. **L.I. Benowitz:** None.

Poster

382. Epilepsy: Networks - Human and Animal Studies

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 382.01/O9

Topic: B.11. Epilepsy

Support: KAKENHI: 15H05874

KAKENHI: 17H05907

Title: Functional connectivity from medial and dorsal parietal areas: A cortico-cortical evoked potential study

Authors: *M. TOGO[1], R. MATSUMOTO[1], T. NAKAE[2], H. TAKEYAMA[3], K. KOBAYASHI[1], K. USAMI[4], A. SHIMOTAKE[5], T. KIKUCHI[2], K. YOSHIDA[2], T. KUNIEDA[6], S. MIYAMOTO[2], R. TAKAHASHI[1], A. IKEDA[5]
[1]Dept. Neurology, Kyoto Univ. Grad. Sch. of Med., Kyoto City, Kyoto, Japan; [2]Dept. of Neurosurg., Kyoto Univ. Hosp., Kyoto-Shi, Japan; [3]Dept. Resp. Care and Sleep Control Med., Kyoto Univ., Kyoto City, Kyoto, Japan; [4]Neurol., Johns Hopkins Univ., Baltimore, MD; [5]Dept. of Epilepsy, Movement Disorders and Physiol., Kyoto Univ. Grad. Sch. of Med., Kyoto, Japan; [6]Neurosurg., Ehime Univ. Grad. Sch. of Med., Toon city, Japan

Abstract: [Objective]The superior parietal lobule (SPL) plays a key role in cognitive processes such as somatosensory and visuomotor, i.e. multimodal integration and visuospatial attention. On the other hand, the medial parietal cortices, especially the precuneus and posterior cingulate cortex belong to default mode network (DMN) being active in resting state. Little is known about connections from the superior and medial parietal cortices in humans. Our objective is to delineate their functional connectivity by means of an electrical, tract-tracing method of cortico-cortical evoked potential (CCEP). [Methods]Subjects were five patients with partial epilepsy and three with a brain tumor, who underwent chronic subdural electrode placement covering the superior or medial parietal cortices (IRB#443). Single-pulse electrical stimuli (1 Hz, pulse width 0.3 msec, 6-12 mA) were delivered to the medial parietal cortex (a total of 35 pairs of electrodes) and the SPL (21 pairs). CCEPs time-locked to the stimuli were obtained by averaging electrocorticograms recorded from subdural electrodes in the frontal (8-72 electrodes/patient), parietal (13-72 electrodes/patient) and occipital (30-39 electrodes/patient) areas. [Results]As for stimulation of the medial parietal area, 1) the precuneus (16 pairs in 6 patients) elicited 66 remote, short-latency CCEPs in the inferior parietal lobule (IPL) (19 remote responses), the SPL (13) and the dorsal premotor area (dorsal PM) (11) as the most active 3 sites. 2) The posterior cingulate cortex (10 pairs in 4 patients) stimulation elicited 36 remote CCEP at the IPL (10), the middle cingulate cortex (5), and the SPL (4). As for the dorsal parietal area, the SPL stimulation (21 pairs in 5 patients) elicited remote 75 CCEPs at the precuneus (17), precentral gyrus (12),

and the dorsal PM (6). [Conclusion]This study implicated i) reciprocal connections between the superior and the medial parietal cortices, and ii) different connectivity pattern to the frontal cortex between the precuneus and the posterior cingulate cortex: the former more with the lateral area, whereas the latter more with the medial area. The findings may explain the mechanism of functional differentiation between these two areas. They also clinically helps us understand seizure propagation network in patients with parietal lobe epilepsy.

Disclosures: **M. Togo:** None. **R. Matsumoto:** Other; Endowed department by UCB, GSK, NihonKoden, Otsuka.. **T. Nakae:** None. **H. Takeyama:** None. **K. Kobayashi:** None. **K. Usami:** None. **A. Shimotake:** None. **T. Kikuchi:** None. **K. Yoshida:** None. **T. Kunieda:** None. **S. Miyamoto:** None. **R. Takahashi:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; DAINIPPON SUMITOMO PHARMA, Boehringer Ingelheim, NOVARTIS, Pfizer Co. Ltd., Takeda Pharmaceutical Company, Mitsubishi Tanabe Pharma, Kyowa Hakko Kirin Co., Otsuka Pharmaceutical Co., Nihon Medi-Physi.. D. Fees for Non-CME Services Received Directly from Commercial Interest or their Agents (e.g., speakers' bureaus); Glaxo Smith Kline, FP PHARMACEUTICAL CORP, NOVARTIS, Boehringer Ingelheim Co., Kyowa Hakko Kirin Co., Dainippon Sumitomo Pharma Co., Mitsubishi Tanabe Pharma Co., Glaxo Smith Kline., Otsuka Pharmaceut. F. Consulting Fees (e.g., advisory boards); KAN Research Institute INC., DAINIPPON SUMITOMO PHARMA, AbbVie Inc., Takeda Pharmaceutical Company. **A. Ikeda:** Other; Endowed department by UCB, GSK, NihonKoden, Otsuka..

Poster

382. Epilepsy: Networks - Human and Animal Studies

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 382.02/DP03/O10 (Dynamic Poster)

Topic: B.11. Epilepsy

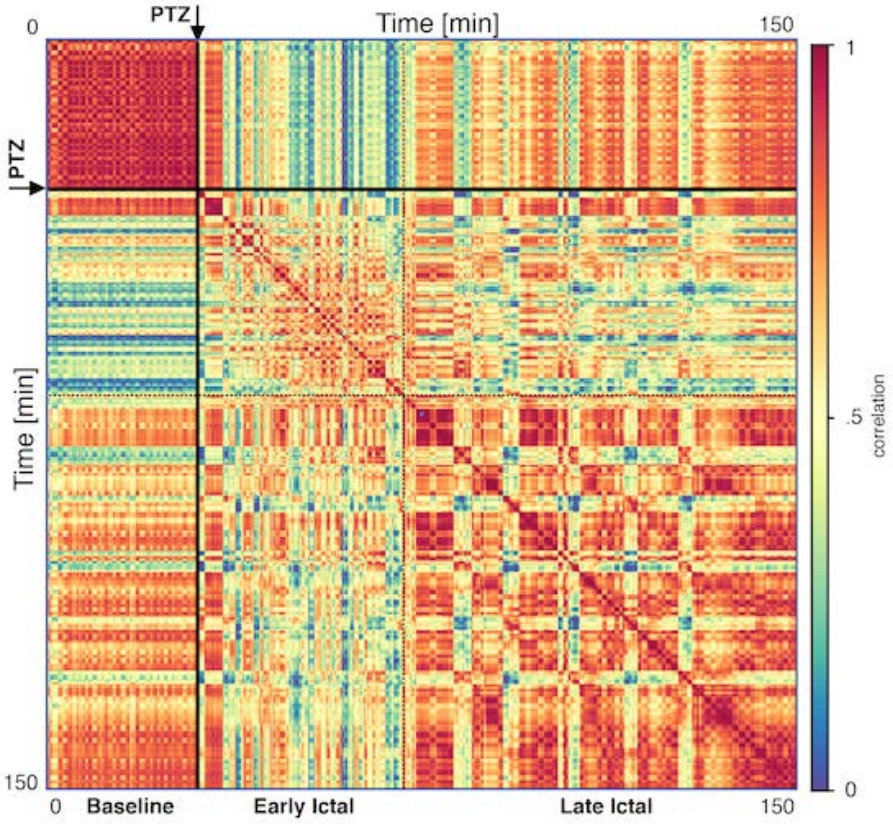
Support: Wellcome Trust Grant 106556/Z/14/Z

Title: Network-wide changes in effective connectivity and synaptic dynamics during epileptic seizures in the larval zebrafish brain

Authors: ***R. E. ROSCH**[1], P. HUNTER[2], T. BALDEWEG[1], K. J. FRISTON[1], M. MEYER[2]
[1]Univ. Col. London, London, United Kingdom; [2]King's Col. London, London, United Kingdom

Abstract: Epilepsy is one of the most common neurological conditions. Pathophysiological explanations typically focus on either the micro/mesoscale (i.e. cortical microcircuitry), or on the macroscale (i.e. functional networks). However, linking abnormalities across different scales remains challenging, partly because of technical limitations in measuring dynamic dysfunction at

a range of observational scales. Recently zebrafish larvae have emerged as a unique model organism to understand epileptic pathophysiology at levels ranging from genetics to whole-organism phenotypes. Furthermore, light sheet calcium microscopy in the larval zebrafish now allows the study of neuronal function in large-scale networks at high temporal resolution in the intact brain *in vivo*. Here we report a computational analysis of functional brain recordings from larval zebrafish during acute epileptic seizures induced with pentylenetetrazole (PTZ).



PTZ causes widespread spectral changes in neuronal activity across all areas. The image above is a dynamic correlation matrix, showing correlation between the distribution of oscillatory power (0-10Hz) across brain areas (optic tectum, midbrain, cerebellum, hindbrain) over time, revealing distinct phases in the seizure. We model these using neural mass models allowing Bayesian inference on the changes in effective network connectivity and synaptic dynamics underlying the neuronal signatures. This dynamic causal modelling (DCM) approach reveals a macroscale shift in network connectivity with a reduction of the influence of higher brain areas during the seizure. Furthermore, seizures are best explained using a combination of changes in inhibitory self-modulatory connections, as well as their synaptic dynamics.
This is consistent with the known mechanism of PTZ, whilst integrating observations into a fully parameterised neural network model of the zebrafish brain. In future work, the detailed single-cell resolution afforded by light-sheet microscopy may be used to validate model prediction made from whole-brain dynamics.

Disclosures: **R.E. Rosch:** None. **P. Hunter:** None. **T. Baldeweg:** None. **K.J. Friston:** None. **M. Meyer:** None.

Poster

382. Epilepsy: Networks - Human and Animal Studies

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 382.03/P1

Topic: B.11. Epilepsy

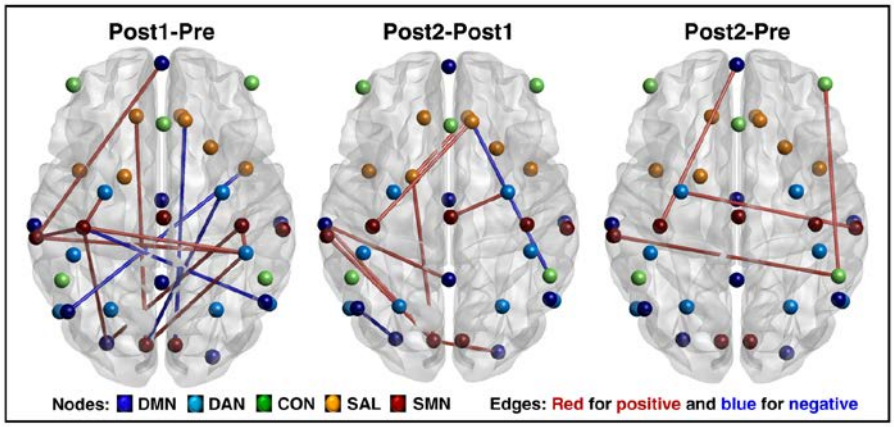
Support: KHIDI grant HI15C2578

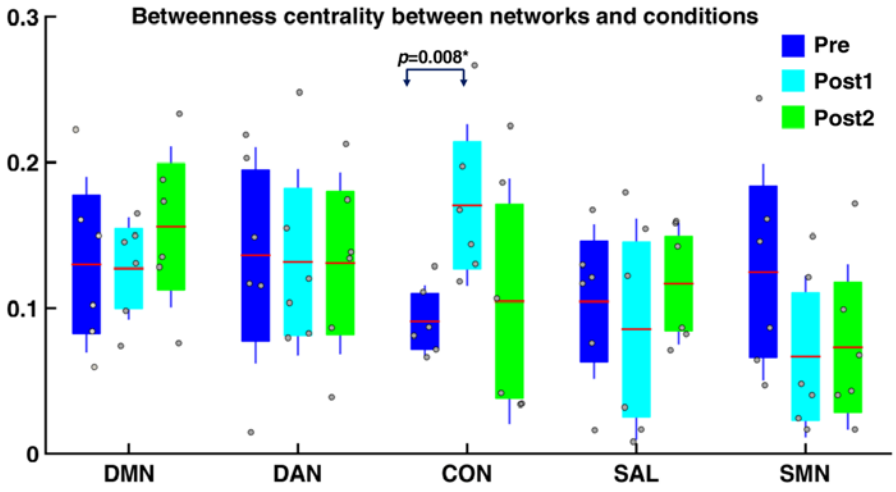
NRF-2017R1A2B4006903

Title: Exercise induced altered functional connectivity in resting state networks in patients with benign epilepsy at alpha band

Authors: G. R. KOIRALA[1], H. KIM[2], N.-Y. KIM[1], *D. LEE[2,3]
[1]Kwangwoon Univ., Seoul, Korea, Republic of; [2]Pediatrics, [3]Epilepsy Res. Inst., Yonsei Univ. Col. of Med., Seoul, Korea, Republic of

Abstract: This study aims to explore the effect of exercise in patients with benign epilepsy through the functional connectivity (FC) measures between 36 cortical region of interests (ROIs) that epitomize five resting state networks: default mode network (DMN), dorsal attention network (DAN), control network (CON), salience network (SAL), and sensorimotor network (SAN). Resting state 19 channel EEG data from six consented patients at three different conditions: pre (before exercise), post1 (after the completion of 5 weeks of supervised as well as home based exercise), and post2 (after the completion of post 1 followed by 30 weeks of home based exercise), were preprocessed and 15 3-s artifact-free epochs were extracted for further analysis. Functional connectivity was evaluated at alpha band (8-13 Hz) using imaginary coherence. Inter-and intra- network alteration in FC was observed following the exercise. Furthermore, post1 betweenness centrality at CON was significantly different from pre-state. The increased connectivity and betweenness centrality after the exercise might be the indication of modulated neuropsychological functions in patients with benign epilepsy.





Disclosures: **G.R. Koirala:** None. **H. Kim:** None. **N. Kim:** None. **D. Lee:** None.

Poster

382. Epilepsy: Networks - Human and Animal Studies

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 382.04/P2

Topic: B.11. Epilepsy

Support: Epilepsy Research UK

Title: Neocortical loss of PGC-1a protects against *In vitro* epileptiform activity

Authors: C. MACKENZIE-GRAY SCOTT[1], R. R. PARRISH[1], R. M. COWELL[2], *C. RACCA[1], A. J. TREVELYAN[1]
[1]Newcastle Univ., Newcastle-upon-Tyne, United Kingdom; [2]Psychiatry, Univ. of Alabama At Birmingham, Birmingham, AL

Abstract: A balance of excitatory and inhibitory transmission is required for proper functioning of the nervous system. Hyperexcitation can cause spontaneous seizures while hypoexcitation can impair learning and memory. Of the interneuron cell types, the parvalbumin-positive (PV+) interneurons exert the greatest control over excitability of pyramidal neuron populations. This is due to their fast-spiking, non-accommodating properties and numerous contacts onto pyramidal neuron somata and axon initial segments. As such, PV+ interneurons are largely responsible for the synchronization of pyramidal neuron networks in the cortex, all being controlled by tight homeostatic regulation of genes and proteins. Alterations in PV+ interneuron function has been associated with epilepsy in humans, and seizures in animal models, possibly due to loss of homeostatic control over gene and protein expression. Evidence from the Cowell lab indicates that the transcriptional coactivator PGC-1α [peroxisome proliferator activated receptor γ (PPARγ) coactivator 1α] is required for the induction of PV in cortical interneurons and normal GABAergic signaling in mice. In addition, the Cowell lab has found that mice lacking PGC-1α, systemically and specifically in PV+ interneurons, exhibit deficiencies in the expression of transcripts involved in synchronous neurotransmitter release. We have found that the initial transcriptional response to seizures involves the induction of PGC-1α and PGC-1α-dependent genes in the neocortex. Since PGC-1α has been shown to influence transmitter release, this suggests that one particular adaptation of PV interneurons to seizure activity is towards more synchronous GABAergic release, thereby promoting synchronicity of cortical networks, which in turn may contribute to further seizure activity and epileptogenesis. In support of this we have found that deletion of PGC-1α in PV+ interneurons delays onset of seizure-like activity in the 0 Mg^{2+} model of epilepsy and decreases overall epileptiform activity. Our RT-PCR studies suggest that this is due to a loss of activity-induced regulation of synaptotagmin 2 (SYT2) and complexin 1 (Cplx1) (molecules involved in synchronous neurotransmitter release), along with loss of PV expression in the PGC-1α knockout mice. We postulate that loss of these genes causes an increase in asynchronous GABA release during evolving epileptiform activity, thereby reducing seizure activity. While we are actively exploring this possibility, these studies open up a set of new transcriptional targets that may be modified to reduce and control seizure severity by altering GABAergic function.

Disclosures: C. Mackenzie-Gray Scott: None. **R.R. Parrish:** None. **R.M. Cowell:** None. **C. Racca:** None. **A.J. Trevelyan:** None.

Poster

382. Epilepsy: Networks - Human and Animal Studies

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 382.05/P3

Topic: B.11. Epilepsy

Support: NIH/NINDS K08 NS069667 (GFB)

NIH/NINDS R01 NS095842 (GFB)

NIH/NINDS T32 NS007421 (BSP)

Beth and Nathan Tross Epilepsy Fund (GFB)

Title: Mechanisms of norepinephrine and serotonin in prevention of seizure-induced respiratory arrest

Authors: *S. W. KRUSE[1], K. G. DAYTON[2], B. S. PURNELL[2], N. K. LEIBOLD[3], G. F. BUCHANAN[2,3]
[1]Univ. of Iowa Col. of Engin., Iowa City, IA; [2]Neurosci. Grad. Program, Univ. of Iowa, Iowa City, IA; [3]Dept. of Neurol., Univ. of Iowa Carver Col. of Med., Iowa City, IA

Abstract: Epilepsy is a highly prevalent disease. About one-third of patients with epilepsy become refractory to medications. Refractory epilepsy patients are at high risk for sudden unexpected death in epilepsy (SUDEP), the leading cause of death in these patients. Seizure-induced respiratory arrest (S-IRA) is an important contributing factor to SUDEP. Serotonin (5-HT) regulates breathing, and 5-HT modulates seizures. A body of circumstantial evidence suggests the 5-HT system may play a critical role in SUDEP, especially in S-IRA. Other neurotransmitters, such as norepinephrine (NE), modulate breathing, affect seizures, and are affected by seizures. Furthermore, NE interacts with 5-HT in many physiological processes. We hypothesized that NE may be involved in regulating breathing following a seizure. Here we examined the effects of pharmacological manipulation of the 5-HT and NE neurotransmitter systems on S-IRA and survival following maximal electroshock (MES)-induced seizures in adult male wild type (WT; *Lmx1b*$^{f/f}$ and C57BL/6) mice, DSP-4 treated NE neuron deficient mice, and transgenic 5-HT neuron deficient (*Lmx1b*$^{f/f/p}$) mice. Mice were implanted with EEG, EMG and EKG electrodes, treated with serotonergic and/or noradrenergic agents, and subjected to MES induced seizures with concomitant respiratory, cardiac and video-EEG monitoring. Seizure severity, mortality, and cardiorespiratory sequelae were assessed. In mice that died, MES induced terminal apnea, while cardiac activity persisted for ≤ 10 min. *Intraperitoneal* (*i.p.)* administration of the norepinephrine reuptake inhibitor (NRI) reboxetine (30 mg/kg) and selective-serotonin reuptake inhibitor (SSRI) fluoxetine (10 mg/kg) 30 min prior to seizure

induction reduced S-IRA in WT and *Lmx1b*$^{f/f/p}$ mice compared to vehicle controls. Application of the α1 receptor antagonist prazosin (10 mg/kg) 15 min prior to SSRI or NRI blocked the protective effects of these agents. The SSRI, citalopram (20 mg/kg) prevented S-IRA in saline treated C57BL/6 mice, but not in C57BL/6 mice subjected to NE neuron destruction with DSP-4 (50 mg/kg x 2). *Intracerebroventricular* administration of the α1 receptor agonist phenylephrine (1 μM) prevented S-IRA and death in WT mice. Taken together, these data suggest NE augmentation with NRI prevents S-IRA and death through an α1-mediated mechanism. That α1 blockade could prevent the protective effects of SSRI, and SSRI was ineffective in mice in which NE neurons were destroyed, suggests that 5-HT and NE interact to modulate breathing after a seizure. Additional work is required to further understand mechanisms of S-IRA and inform prophylactic strategies for SUDEP.

Disclosures: S.W. Kruse: None. **K.G. Dayton:** None. **B.S. Purnell:** None. **N.K. Leibold:** None. **G.F. Buchanan:** None.

Poster

382. Epilepsy: Networks - Human and Animal Studies

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 382.06/P4

Topic: B.11. Epilepsy

Support: NIH Grant R01MH100186

Title: Maturation of motor cortex excitability in children with focal epilepsy as measured by navigated transcranial magnetic stimulation

Authors: *H. L. KAYE[1], G. BLOCK[4], A. JANNATI[4], C. VEGA, III[2], K. RAKESH[2], K. KAPUR[2], L. M. OBERMAN[5], R. GERSNER[2], A. BOES[6], A. PASCUAL-LEONE[4], A. ROTENBERG[3,4]
[1]Dept. of Neurology, Div. of Epilepsy and Clin. Neurophysiol., [2]Neurol., [3]Boston Children's Hosp., Boston, MA; [4]Neurology, Ctr. Noninvasive Brain Stimulation, Beth Israel Deaconess Med. Ctr., Boston, MA; [5]Bradley Hosp., Riverside, RI; [6]Departments of Pediatrics, Neurol. & Psychiatry, Univ. of Iowa, Iowa City, IA

Abstract: Objectives
(1) To measure the developmental trajectory of corticospinal tract (CST) excitability as a function of hemispheric lateralization and proximity to seizure focus in children with epilepsy, undergoing navigated transcranial magnetic stimulation (nTMS); (2) To test the relationship between CST excitability and patient IQ.
Methods
137 patients (≤18 y) with intractable focal epilepsy underwent nTMS for functional motor

mapping to measure bilateral resting motor threshold (rMT). Only patients with rMT <100% machine output were included for analysis. Comparisons between healthy and epileptic hemispheres were performed by a linear mixed-effects model to account for within-subject correlations, with age, lesion, and age x lesion interaction as fixed-effects. Further analysis was performed using mixture modeling to detect clustering of rMT with respect to age per hemisphere. Bayesian Information Criterion was used to choose number of clusters in the mixture models. Multiple regression analyses examined the relationship between rMT and verbal IQ, while controlling for age, for the clusters identified above.

Results

In patients without CST lesion there is no rMT difference between epileptic and healthy hemispheres. Age is the major determinant of rMT, which decreases by ~7 V/m per year in childhood (p<.001); this maturational trajectory does not differ between hemispheres. Cluster analysis reveals two groups corresponding to (1) mature, minimal rMT after age ~15 y and (2) a progressive maturation from age 7 to 15 y. In a subgroup of patients (n=14) with CST injury in the epileptic hemisphere, average rMT is lower in the epileptic hemisphere (p<.01). In this group, rMT decreases by ~9 V/m per year in the healthy hemisphere (R^2 = .57; p<.01), but does not change with age in the epileptic hemisphere (R^2=.005; p=.82). The difference in maturational trajectory is significant between the two hemispheres (p<.01). After controlling for age, healthy hemisphere rMT correlates negatively with verbal IQ in the age □15 y group (R^2=.39; p=.036), but not in the age 7-15 y group. Epileptic hemisphere rMT is not correlated with verbal IQ in either group (p's > .44).

Conclusions

Cortical excitability increases with age until ~ age 15 yrs. We thus describe for the first time the rate and endpoint of CST maturation. Neither rate of maturation nor absolute rMT differ between healthy and epileptic hemispheres in patients with focal epilepsies without CST injury. However, in patients with a unilateral CST lesion, the maturational trajectory is absent in the injured hemisphere. We also identify a positive correlation between rMT and verbal IQ in patients with mature CST excitability.

Disclosures: H.L. Kaye: None. **G. Block:** None. **A. Jannati:** None. **C. Vega:** None. **K. Rakesh:** None. **K. Kapur:** None. **L.M. Oberman:** None. **R. Gersner:** None. **A. Boes:** None. **A. Pascual-Leone:** F. Consulting Fees (e.g., advisory boards); Magstim, Nexstim, Neronix,Neuroelectrics, Starlab Neuroscience, Constant Therapy, Neosync, axilum robotics. **A. Rotenberg:** F. Consulting Fees (e.g., advisory boards); Neuro'motion Inc., consults for NeuroRex Inc., and a co-inventor of a patent for real-time integration TMS and EEG. AR receives or has received research support in the form of material and/or funding.

Poster

382. Epilepsy: Networks - Human and Animal Studies

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 382.07/P5

Topic: B.11. Epilepsy

Title: Human induced pluripotent stem cell (hiPSC) -derived neurons respond to convulsant drugs when co-cultured with hiPSC-derived astrocytes

Authors: *M. N. ISHII[1], K. YAMAMOTO[2], M. SHOJI[2], A. ASAMI[2], Y. KAWAMATA[2]
[1]CNS Drug Discovery Unit, Takeda Pharmaceut. Co. Limited, Fujisawa, Kanagawa, Japan; [2]Takeda Pharmaceut. Co. Limited, Kanagawa, Japan

Abstract: Accurate risk assessment for drug-induced seizure is expected to be performed before entering clinical studies because of its severity and fatal damage to drug development. Induced pluripotent stem cell (iPSC) technology has allowed the use of human neurons and glial cells in toxicology studies. Recently, several studies showed the advantage of co-culture system of human iPSC (hiPSC)-derived neurons with rodent/human primary astrocytes regarding neuronal functions. However, the application of hiPSC-derived neurons for seizure risk assessment has not yet been fully addressed, and not at all when co-cultured with hiPSC-derived astrocytes. Here, we characterized hiPSC-derived neurons co-cultured with hiPSC-derived astrocytes to discuss how hiPSC-derived neurons are useful to assess seizure risk of drugs. First, we detected the frequency of spikes and synchronized bursts hiPSC-derived neurons when co-cultured with hiPSC-derived astrocytes for 8 weeks. This synchronized burst was suppressed by the treatment with 6-cyano-7-nitroquinoxaline-2,3-dione, α-amino-3-hydroxy-5-methylisoxazole-4-propionic acid (AMPA) receptor antagonist, and D-(-)-2-amino-5-phosphonopentanoic acid, an N-Methyl-D-aspartate (NMDA) receptor antagonist. These data suggested that co-cultured hiPSC-derived neurons formed synaptic connections mediated by AMPA and NMDA receptors. We also demonstrated that co-cultured hiPSC-derived neurons showed epileptiform activity upon treatment with gabazine or kaliotoxin. Finally, we performed single-cell transcriptome analysis in hiPSC-derived neurons and found that hiPSC-derived astrocytes activated the pathways involved in the activities of AMPA and NMDA receptor functions, neuronal polarity, and axon guidance in hiPSC-derived neurons. These data suggested that hiPSC-derived astrocytes promoted the development of action potential, synaptic functions, and neuronal networks in hiPSC-derived neurons, and then these functional alterations result in the epileptiform activity in response to convulsant drugs. Our study indicates the possibility that co-culture system of hiPSC-derived neurons with astrocytes could be useful in the risk assessment of drug-induced seizure.

Disclosures: **M.N. Ishii:** None. **K. Yamamoto:** None. **M. Shoji:** None. **A. Asami:** None. **Y. Kawamata:** None.

Poster

382. Epilepsy: Networks - Human and Animal Studies

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 382.08/P6

Topic: B.11. Epilepsy

Support: 2017 SfN-IBRO travel award

CNPQ

Title: Alterations in pre-ictal oscillation of pilocarpine model of epilepsy and the effects of MGE-grafted precursor cells

Authors: *M. VENDRAMIN PASQUETTI[1], S. A. A. ROMARIZ[2], Í. F. MESSIAS[1], B. MONTEIRO LONGO[2], M. E. CALCAGNOTTO[1]
[1]Univ. Federal Do Rio Grande Do Sul, Porto Alegre, Brazil; [2]Univ. Federal de São Paulo, São Paulo, Brazil

Abstract: Introduction: GABAergic interneurons are crucial for local circuitry and have major role in epilepsy. Most of GABAergic interneurons originates from medial ganglionic eminence (MGE). MGE-derived cells grafted into brain of animal models of epilepsy migrate, differentiate into mature GABAergic cells, recover inhibition, restore brain rhythms and reduce spontaneous recurrent seizures (SRS). However, their effect on pre-ictal oscillation at seizure onset (SO) has not been explored. Methods: We analyzed pre-ictal pattern oscillations in pilocarpine model of epilepsy. Seven days after Pilocarpine induced *Status epilepticus*, rats received or not MGE grafts into hippocampus. The animals were video-monitored (9h/d-90d) and reduction of SRS frequency was observed in MGE-Pilo group. Later, cortical and intrahippocampal electrodes were implanted for video-EEG recording (24h/d-7d). The EEG data were decomposed by Matlab, in delta:1-4Hz, theta:4-12Hz, low gamma:20-50Hz, high gamma:60-100Hz and ripples:100-200Hz oscillations at 5min and 20s before SO. Ethical committee approval: 0024/12. Results/Discussion: Both groups had a decrease in cortical and hippocampal delta and theta and hippocampal low gamma waves 20s before SO. Cortical high gamma decrease in Pilo animals, but not in MGE-Pilo. Ripples increased in both structures in MGE-Pilo group. Theta/delta ratio decreased in both groups in cortex, but in hippocampus that happen only in MGE-Pilo group. That could contribute to reduction in SRS frequency observed in the transplanted animals. Understanding how changes in neuronal circuitry tuned brain oscillation before SO may have predictive and therapeutic value.

Disclosures: **M. Vendramin Pasquetti:** None. **S.A.A. Romariz:** None. **Í.F. Messias:** None. **B. Monteiro Longo:** None. **M.E. Calcagnotto:** None.

Poster

382. Epilepsy: Networks - Human and Animal Studies

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 382.09/P7

Topic: B.11. Epilepsy

Support: NIH R01 NS034700

Title: Disruption of seizures with high resolution targeted optical stimulation *In vitro*

Authors: *K. P. LILLIS[1,2], K. J. STALEY[1,2]
[1]Neurol., Harvard Med. Sch. Dept. of Neurol., Charlestown, MA; [2]Neurol., Massachusetts Gen. Hosp., Boston, MA

Abstract: Currently available anti-seizure therapies broadly affect activity in the brain by altering ion channels/neurotransmission (drugs), metabolic processes (diet), or regional activity (electrical stimulation). The non-specific nature of these treatments naturally results in undesirable neurological side effects. In this work we are determining the minimum intervention that is sufficient to prevent ictogenesis, using targeted optogenetic activation of individual neurons.

Organotypic hippocampal slice cultures, which generate spontaneous periodic seizures after approximately one week in culture, were prepared from animals transduced with AAV vectors coding for the light-sensitive, high-current channelrhodopsin CoChR and the red-shifted calcium indicator jRCaMP1a. When imaging relatively thin samples with sparse expression, it is possible to resolve activity in individual neurons using standard single photon widefield microscopy with a low-magnification objective lens, whose field-of-view spans the entire slice culture. Furthermore, we incorporated a digital micromirror device (DMD) in the conjugate image plane of the blue excitation light pathway, which enabled projection of high resolution patterns onto the slice. By co-registering the camera and DMD, we could target any subset of individually imaged neurons for optical stimulation.

While optically monitoring seizure activity using the jRCaMP1a signal, we disrupted activity by periodically stimulating neurons in one of three hippocampal subregions: *s. radiatum, s. pyramidale,* or *s. oriens*. For each region 360 selected cells were divided into 6 groups of 60 cells. The 60-cell patterns were cycled through at 6 Hz, resulting in 1Hz stimulation per targeted cell. Preliminary data show that stimulation of cells in s.p. reliably prevents seizures for > 30 minutes in slices that were undergoing 60 second seizures at every 3 minutes at baseline. Immediately after turning off the stimulation light, slices returned to their baseline seizure pattern. Stimulation of s.o. and s.r. were less effective, preventing seizures for a maximum of 15

minutes before seizures emerged despite ongoing optical stimulation. We are characterizing the impact of different stimulation strategies on spontaneous network activity, and exploring the vast stimulation parameter space to identify the smallest population of neurons whose manipulation prevents seizures, with minimal disruption of baseline activity.

Disclosures: K.P. Lillis: None. **K.J. Staley:** None.

Poster

382. Epilepsy: Networks - Human and Animal Studies

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 382.10/P8

Topic: B.11. Epilepsy

Support: U54 OD020351

R01 NS057482

Schaefer Scholarship, Columbia University

Title: Stress testing cortical networks: Using simple pharmacological challenges to understand the effects of genetic mutations on cortical network excitability

Authors: N. CODADU[1], R. R. PARRISH[2], D. C. DE VIVO[3], M. TANG[4], C. A. SCHEVON[4], U. MONANI[5], W. N. FRANKEL[6], *A. J. TREVELYAN[1]
[1]Newcastle Univ., Newcastle Upon Tyne, United Kingdom; [2]Inst. of Neurosci., Univ. of Newcastle, Newcastle Upon Tyne, United Kingdom; [3]Dept Neurol., Columbia Univ. CPS, New York, NY; [5]MNC, [4]Columbia Univ., New York, NY; [6]Columbia Univ. Med. Ctr., New York, NY

Abstract: Bathing brain slices in modified cerebro-spinal fluid (CSF) has proved a useful experimental tool for understanding epileptic activity. The two most commonly used modifications are either to remove Mg^{2+} ions from the bathing medium, or to add the K channel blocker, 4-aminopyridine (4AP). These two manipulations however, induce activity that evolves in very different and highly characteristic ways. As such, these simple pharmacological models provide a wide range of metrics, many of which can be related to specific components of the network. Rather than using these models to examine the endstage, full ictal activation, instead, we use them to “stress test” cortical preparations from different transgenic animals. We will present data from a range of wild-type and transgenic mouse lines to illustrate how these studies can expand our understanding of the effects of specific mutations on cortical network function. Horizontal brain slices, including the hippocampal, entorhinal and neocortical territories, were prepared from young adult mice, and evolving network activity was recorded extracellularly at multiple cortical locations, as the tissue was perfused with either 0 Mg or 4AP CSF in an

interface chamber. Our initial explorations of wild-type animals showed that the results were sensitive to the animal age and preparation protocol, highlighting the importance of using age-matched, littermate, wild-type controls for comparison with the recordings of transgenic animals. Recordings and the initial analyses of the transgenic / wild-type animals were made blind to the genotype. Brain slices were laid over a 96 microelectrode array (400µm separation, Blackrock Microsystems), to provide a dense sampling of all cortical regions. We will present our initial analyses of 3 different transgenic lines, carrying mutations in the Dnm1 and Glut1 genes. Both these mutations have been associated with severe epileptic phenotypes in clinical cases.
This work was funded by a Schaefer Scholarship (AJT), and NIH awards R01 NS057482 (UM) and U54 OD020351 (WF).

Disclosures: N. Codadu: None. **R.R. Parrish:** None. **D.C. De Vivo:** None. **M. Tang:** None. **C.A. Schevon:** None. **U. Monani:** None. **W.N. Frankel:** None. **A.J. Trevelyan:** None.

Poster

382. Epilepsy: Networks - Human and Animal Studies

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 382.11/P9

Topic: B.11. Epilepsy

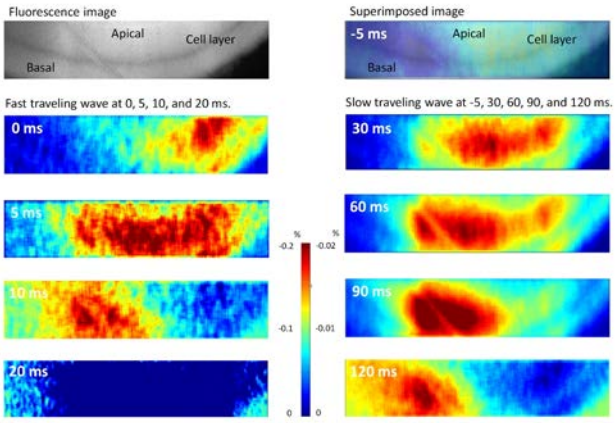
Support: NIH Grant 5R01NS060757

Title: In-vitro slow moving focus in the epileptic hippocampus mimics propagation of human seizures

Authors: *C.-C. CHIANG[1], X. WEI[2], A. ANANTHAKRISHNAN[2], D. M. DURAND[3]
[1]Biomed. Engin., [2]Case Western Reserve Univ., Cleveland, OH; [3]Dept Biomed. Eng, Case Western Res. Univ., Cleveland, OH

Abstract: Fast and slow neural waves have been observed to propagate in the human brain during seizures using recording arrays. Yet the nature of these waves is difficult to determine in a surgical setting. Here we report the observation of fast and slow waves propagate in the hippocampus in-vitro at speeds similar to those in human tissue. The fast and slow waves were recorded with a genetically encoded voltage sensitive fluorescent protein (VSFP Butterfly 1.2) and a high speed camera. The results of this study indicate that the fast wave is NMDA-sensitive but does not require synaptic transmission for propagation. Image analysis indicates that the slow wave is the source of the fast wave and is therefore a moving focus of the seizure-like activity (see Figure). This slow moving focus is associated with a propagating neural calcium wave detected with calcium dye (OGB-1) but is independent of NMDA receptors, not related to ATP release, and cannot be explained by potassium diffusion. Computer simulation suggests that the

slow moving focus is a calcium spike and propagates by ephaptic coupling. These results indicate that a seizure-like focus can propagate non-synaptically and provide an alternative explanation for seizure progression in the brain.



Disclosures: **C. Chiang:** None. **X. Wei:** None. **A. Ananthakrishnan:** None. **D.M. Durand:** None.

Poster

382. Epilepsy: Networks - Human and Animal Studies

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 382.12/P10

Topic: B.11. Epilepsy

Support: NIH/NINDS R01 NS069861 (V.S)

NJCBIR CBIR14RG024 (V.S)

Title: Distinct inhibitory regulation of dentate granule and semilunar granule cells

Authors: *M. AFRASIABI, V. SANTHAKUMAR
Pharmacology, Physiol. and Neurosci., Rutgers, New Jersey Med. Sch., Newark, NJ

Abstract: The dentate gyrus has been proposed to serve as a functional “gate” that regulates inputs into the hippocampus. Apart from the classical dentate projection neurons, Granule Cells (GCs), recent studies have identified a new group of excitatory neurons, Semilunar Granule Cells (SGCs). SGCs support feedback inhibition of GCs and thereby maintain the dentate gate. We previously reported that SGCs receive higher frequency of spontaneous inhibitory post-synaptic currents (sIPSCs) than GCs, and unlike GCs, show reduced sIPSC frequency after brain injury,

indicating distinctive inhibitory regulation of the two cell types. Based on their morphology, we propose that SGCs are not under the strong gating feedback inhibitory regulation as GCs. Here we examined whether parvalbumin-expressing presumed fast-spiking interneurons (PV-IN) and GABAergic neurons expressing the cannabinoid receptor type 1 (CB1R) have differential contributions to basal and evoked synaptic inhibition in SGCs and GCs. Consistent with data from rats, whole cell recordings and biocytin immunostaining in hippocampal sections from mice showed that SGCs can be distinguished from GCs by somato-dendritic structure and intrinsic physiology. SGCs receive significantly higher sIPSCs than GCs and show sustained firing upon perforant path (PP) stimulation. In contrast, PP activation resulted in a greater and more sustained increase in IPSCs in GCs compared to SGCs. Optogenetic silencing of PV-INs failed to reduce sIPSC frequency in SGCs while optogenetic activation of them resulted in a greater increase in sIPSC frequency in GCs than in SGCs (GC: 719.13±141.61%, n=14, SGC: 355.59±40.16%, n=14, p=0.02 by t-test). WIN-2 (a CB1R agonist) failed to reduce baseline sIPSC frequency in both cell types showing that CB1R sensitive interneurons, which include dentate CCK-expressing and total molecular layer (TML) interneurons, have limited contribution to baseline sIPSC frequency in GCs or SGCs. Our data demonstrate that SGC have limited baseline and evoked inhibition from PV interneurons and may elude the robust dentate inhibitory gate.

Disclosures: M. Afrasiabi: None. **V. Santhakumar:** None.

Poster

382. Epilepsy: Networks - Human and Animal Studies

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 382.13/Q1

Topic: B.11. Epilepsy

Support: NIH Grant R21 NS088608

NIH Grant R01 DK056132

Title: Reduced voltage-gated K^+ channel function in gabaergic nts neurons in a murine model of acquired TLE and SUDEP

Authors: *I. DERERA[1], B. N. SMITH[1,2]
[1]Dept. of Physiol., [2]Epilepsy Ctr., Univ. of Kentucky, Lexington, KY

Abstract: Sudden unexpected death in epilepsy (SUDEP) accounts for approximately 17% of epilepsy-related deaths. Altered voltage-gated K^+ channel function in neurons of the brainstem nucleus tractus solitarius (NTS) and vagal afferent fibers may contribute to SUDEP in genetic epilepsy models; hippocampal voltage-gated K^+ channel function and expression is altered in the

pilocarpine-induced status epilepticus (SE) model of acquired temporal lobe epilepsy (TLE) in rodents. However, little is known regarding possible changes in voltage-gated K^+ channels in NTS neurons during development of acquired TLE. GABAergic NTS neurons receive information via vagal afferent fibers regarding cardiac and respiratory function and serve to integrate, filter, and modulate this information to regulate cardiorespiratory function. In a genetic epilepsy model with a voltage-gated K^+ channel mutation, altered NTS neuron function contributed to cardiorespiratory collapse and sudden death after seizures. However, there is a paucity of information regarding epilepsy-related changes in NTS neuron channel function in acquired epilepsy, which affects a greater proportion of the patient population. We hypothesized that voltage-gated K^+ channel function in GABAergic NTS neurons is altered in the pilocarpine-induced SE model of TLE. Pilocarpine (282 mg/kg) was administered to 4 week old male mice that express GFP in a subset of GABAergic NTS neurons (FVB-Tg(GADGFP)4570Swn/J; ie. GIN mice) to induce SE and eventual development of TLE. *In vitro* electrophysiological results show an increase in action potential firing rate and action potential half-width in GABAergic NTS neurons from mice 12 weeks after SE (i.e., TLE mice) compared to age-matched controls. Upon application of 4-AP (5mM), action potential firing rate and half-width in GABAergic NTS neurons from control mice was increased to levels similar to that in neurons from TLE mice, suggesting that A-type K^+ current function may be suppressed following TLE development. Characteristics of the A-type K^+ current were therefore assessed in voltage-clamp recordings in GABAergic NTS neurons from control and TLE mice. Preliminary data suggest that peak A-type K^+ current is decreased in GABAergic NTS neurons from TLE mice compared to controls, consistent with the increase in action potential firing and half-width in TLE mice. These results suggest voltage-gated K^+ channel function is reduced in the NTS of mice with acquired TLE, which contributes to increased neuronal activity and may also increase SUDEP risk.

Disclosures: I. Derera: None. **B.N. Smith:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.01/Q2

Topic: B.11. Epilepsy

Support: CONACYT-BMBF 2013 Grant Number 208132

Beca de Posgrado Conacyt, Doctorado en ciencias Biomédicas, UNAM

Title: Seizure characterization and electroencephalographic features in TLR379 triple knockout mice

Authors: *N. SALEH-SUBAIE[1], O. ARIAS-CARRION[2]
[1]UNAM, México City, Mexico; [2]Trastornos Del Movimiento Y Sueño (TMS), Hosp., MEXICO CITY, Mexico

Abstract: Epilepsy is a disabling neurological disorder characterized by recurring, unprovoked seizures and affects about 1% of the population. It involves several processes, such as neuronal loss, neurogenesis, and inflammation. The role of inflammatory mediators such as Toll-like receptors (TLRs) is under review. TLRs were involved in several neuronal disorders like Parkinson and Epilepsy. Nevertheless, the roles of TLRs in the CNS, are still poorly understood. In a recent study, it has been demonstrated that TLR 3 induces expression of a range of neuroprotective mediators such as interleukin-9 (IL9), IL10 and IL11, moreover, several other molecules that regulate cellular growth, differentiation, and migration. These results demonstrated that the expression of TLR3 in human brain slice cultures mediates a neuroprotective response in a pro-inflammatory reaction. Most recently, a role of TLR9 contributing to the neuronal protection has been described by used a model of epilepsy in mice (kainic acid). However few studies link the receptors in epilepsy. The inflammatory mechanisms that are involved with the disease are still not fully elucidated. It was published that TLR379 triple knockout mice showed that in spleen and thymus the mice express strong amounts of uncontrolled endogenous retroviruses. Furthermore, those mice showed spontaneous epileptic seizures. This suggests that enhanced activity of TLR 3, 7, or 9 could be used to treat epilepsy, as they may reduce seizure frequency. For that reason, the aim of this project was to characterize the electrographic and behavioral activity in TLR379 triple knockout mice. TLR379 triple knockout mice were on a C57BL/6 background. For a first approach, we performed cFos immunostaining one hour after an event that exhibited behavioral characteristics corresponding with stage 5 of the Racine seizure scale. We also analyzed the brains for abnormalities and cellular density (Nissl staining). To characterize, the electrographic activity mice were implanted with a unipolar electrode into the left ventral hippocampus and were registered up to seven days. The electrographic activity of hippocampus was analyzed using fast Fourier transform method throughout all the experiment. The animals were sacrificed and the brains used to evaluate the site of electrode implantation (Nissl staining). To assess the behavioral changes, in spatial memory acquisition we performed a water maze tests. We found seizure activity and epileptiform activity in TLR379 triple knockout mice and significant changes in spatial memory acquisition as well compare with our control group.
Support: CONACYT-BMBF 2013 Grant Number 208132

Disclosures: **N. Saleh-Subaie:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CONACYT-BMBF 2013 Grant Number 208132. **O. Arias-Carrion:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CONACYT-BMBF 2013 Grant Number 208132.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.02/Q3

Topic: B.11. Epilepsy

Support: NRF Korea Grant 2015R1C1A1A01052351

Title: Calpain/STEP pathway contributes prenatal stress-induced epileptic spasms in infant rat

Authors: H. KWON[1], J. HONG[2], D. KIM[4], *J. KANG[3]
[1]Med. science, Chungnam Natl. Univ. Sch. of Med., Daejeon, Korea, Republic of; [3]Pediatrics, [2]Chungnam Natl. Univ., Daejeon, Korea, Republic of; [4]Anat., Chungnam Natl. Univ. Sch. Med., Daejeon, Korea, Republic of

Abstract: Infantile spasms (IS) is a serious epileptic syndrome that occurs in the early infantile age. Recent studies revealed prenatal stress exposure is a risk factor of IS. Our previous study showed that prenatal stress with betamethasone altered the maturation of GABAergic progenitors and resulted in the lack of GABA input, which in turn, decreased KCC2 expression and lowered the seizure threshold. However, the mechanisms on the seizure susceptibility to N-methyl-D-aspartate (NMDA)-triggered spasms on postnatal day still need to be clarified. Prenatal stress with betamethasone on gestational day 15 increased seizure susceptibility to NMDA-triggered spasms on postnatal day 15. Compared to the control group, the protein level of NMDA receptor 2B (NR2B) was not different, but the protein level of phosphorylated-NR2B/NR2B ratio was decreased in the cortex of the prenatally stressed model. We further confirmed increased expression levels of STEP, phosphorylation of p38, cleaved caspase-3, and decreased calpain2. Our study suggest that calpain2 and STEP pathway and downstream p38 are related with increased sensitivity of seizure threshold of prenatal stress-induced cortex, and it would be considered as a new treatment approach of infantile spasms.

Disclosures: H. Kwon: None. **J. Hong:** None. **D. Kim:** None. **J. Kang:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.03/Q4

Topic: B.11. Epilepsy

Support: Epilepsy Foundation of America

Citizens United for Research in Epilepsy

Nebraska State LB692

NINDS NS072179

Title: Pharmacoresponsivness of seizures and sleep-wake patterns of Kcna1-null mice, a model of temporal lobe epilepsy with co-morbid sleep disorders to conventional anti-seizure drugs (ASDs)

Authors: *M. DEODHAR[1], B. THOMAS[2], L. ADAMIAN[3], S. MATTHEWS[3], K. A. SIMEONE[4], T. A. SIMEONE[5]
[1]CREIGHTON UNIVERSITY, Omaha, NE; [2]Creighton UNiversity, Omaha, NE; [3]Creighton Univ., Omaha, NE; [4]Dept. of Pharmacol., [5]Pharmacol., Creighton Univ. Sch. of Med., Omaha, NE

Abstract: Objective: Thirty percent of patients affected with epilepsy are resistant to conventional ASDs. Kcna-1 null mice are well recognized as a model for temporal lobe epilepsy and more recently for SUDEP. These mice exhibit severe generalized tonic clonic seizures, extensive neuropathology in the brain, and also a co-morbid sleep disorder, all of which are risk factors for refractory epilepsy. Recently, it has been demonstrated that Kcna1-null mice respond to dietary and investigational therapies; however, whether Kcna1-null mice respond to conventional drugs is unknown. Here, we aimed to determine the effects of the conventional ASDs phenobarbital, carbamezapine, phenytoin and levetiracetam, on seizures and sleep-wake patterns in the Kcna-1 null mice. Based on the risk factors exhibited, we hypothesized that Kcna1-null mice will not respond to multiple ASDs.
Methods: Kcna-1 null littermates were housed in a 12 hr light-dark cycle with access to food and water *ad libitum*. At P27-29, subdural EEG electrodes were implanted and a head-mount was attached to the mice for EEG recordings. Following a seven-day recovery period, mice were i.p. injected with vehicle once daily for two consecutive days, followed by two consecutive days of once daily injections of the ASD. Continuous video-EEG recordings were carried out throughout this period. Seizure activity and sleep-wake was analyzed for all 96 hours of recording per animal using Sirenia and Spike2 software.
Results: Phenobarbital eliminated seizures for 24 hours post-injection and significantly increased sleep duration when compared to the vehicle. Phenytoin caused an initial non-significant reduction in seizures but then returned back to baseline after 4 hours. Carbamezapine caused an initial non-significant reduction followed by an exacerbation in seizures after 4 hours. Phenytoin and carbamezapine both caused non-significant increases in sleep duration. Analyses for effects of levetiracetam are ongoing.
Conclusions: Kcna1-null mice were responsive to the GABAA receptor modulator phenobarbital and unresponsive to the sodium channel blockers phenytoin and carbamazepine suggesting that Kcna1-null mice can be tentatively labeled as a model of refractory epilepsy. Whether this

refractoriness is limited to ASDs that have sodium channel inhibition as primary mechanisms of action requires further studies.

Disclosures: **M. Deodhar:** None. **B. Thomas:** None. **L. Adamian:** None. **S. Matthews:** None. **K.A. Simeone:** None. **T.A. Simeone:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.04/Q5

Topic: B.11. Epilepsy

Support: NIH Grant F040383

Title: Temporal dynamics of high-frequency oscillations in a pilocarpine rat model using a novel automated detector

Authors: *J. SCOTT, S. REN, S. GLISKE, H. LUNA-MUNGUIA, W. STACEY
Univ. of Michigan, Ann Arbor, MI

Abstract: Most automated detectors of high-frequency oscillations (HFO) in humans are designed for interictal time periods, and exclude other epochs (e.g. ictal, preictal) from detection. However, our prior work has shown that HFO morphology evolves temporally as seizure onset is reached. Further investigation into these dynamics therefore necessitates the development of an automated HFO detector capable of continuous detection across all epochs. Optimization of translational epilepsy therapies requires that any automated detector function properly within the constraints of an animal model. Here we present a continuous rat HFO detector, and with it an analysis of HFOs recorded intracranially across all time epochs in a pilocarpine rat model of temporal lobe epilepsy. Using the detector, we find statistically significant increased median HFO rates in epileptic animals (n=16) over controls (n=11). Utilizing unsupervised learning methods such as PCA, we find further that feature distributions of preictal HFOs differ significantly from those at baseline within individuals. Our findings suggest that evolving HFO dynamics play a role in seizure generation. By exploiting these changes, future work could lead to more effective patient-specific treatment modalities - such as more accurate seizure prediction algorithms, or even adaptive closed-loop neural stimulation systems using HFOs as an input control signal.

Disclosures: **J. Scott:** None. **S. Ren:** None. **S. Gliske:** None. **H. Luna-Munguia:** None. **W. Stacey:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.05/Q6

Topic: B.11. Epilepsy

Support: Bill and Melinda Gates Foundation Cysticercosis Elimination in Peru grants 23981 and 33848 (H.H.G.)

NIH grant 5D43TW006581 (Infectious Diseases Training Program in Peru) (R.H.G.)

NIH training grant FIC/NIH D43 TW001140, and Fondo para la Innovación, Ciencia y Tecnología FINCyT grant 129-FINCyT-IB-2013 (M.R.V.)

Innovate Perú Nro.135-PNICP-PIAP-2015

Title: Change in electroencephalography pattern in a rat model of Neurocysticercosis

Authors: *A. D. DELGADO[1], R. P. CARMEN OROZCO[1], R. H. GILMAN[2], L. E. BAQUEDANO[1], D. G. DÁVILA VILLACORTA[1], G. CASTILLO[1], J. MORALES[1], N. CHILE[1], R. H. CELÍZ[1], M. R. VERASTEGUI[1]
[1]Infectious Dis. Lab. Research-LID, Univ. Peruana Cayetano Heredia, Lima, Peru; [2]Intl. Health, Bloomberg Sch. of Hyg. and Publ. Hlth., Johns Hopkins Univ., Baltimore, MD

Abstract: Neurocysticercosis (NCC) is caused by Taenia solium larva stage, infecting the central nervous system. It is the principal responsable of late-onset epilepsy in endemic countries. Our group has development a model rat to understand the physiology for neurocysticercosis allowing us to describe the EEG waveforms of seizures. This novel model offers the development of viable cysticerci and the main clinical manifestation of the disease, which is epilepsy, making the development of the disease in the rat model similar than in humans. In a preliminare study we observed that 9% of the rats after 5 months of infection, develop generalized clonic-tonic seizures. Our goal is to characterizate of the electroencephalogram record generated by neurocysticercosis in a rat model, and to determinate if the pattern in the EEG register is associated with spontaneous seizures. Male Holtzman rats received intracranial infection with activated T. solium oncospheres between 12-15 days of birth, MRI T2 were performed in order to detect the presence of the cysticercus in the rat brain, after 3 months of infection. Selected Infected rats (n=10) and not infected rats (n=5), implanted with cortical electrodes, were continuously recorded by telemetric electroencephalography (tEEG) to monitor the brain activity and detect spontaneous seizures for five weeks. We found variations in the electroencephalographic record in a rat model of neurocysticercosis, with a pattern of sharp

waves and spikes, spontaneous seizures were observed in all infected rats, varying the frequency and number of seizures per rat.

Disclosures: **A.D. Delgado:** None. **R.P. Carmen Orozco:** None. **R.H. Gilman:** None. **L.E. Baquedano:** None. **D.G. Dávila Villacorta:** None. **G. Castillo:** None. **J. Morales:** None. **N. Chile:** None. **R.H. Celíz:** None. **M.R. Verastegui:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.06/Q7

Topic: B.11. Epilepsy

Support: CIHR grant M101111

Title: Excess GIRK2 activity is necessary but not sufficient to confer susceptibility to Infantile Spasms

Authors: *K. JOSHI[1], L. SHEN[2], M. A. CORTEZ[2], O. SNEAD[2]
[1]The Hosp. For Sick Children, Toronto, ON, Canada; [2]Hosp. for Sick Children, Toronto, ON, Canada

Abstract: Infantile spasms (IS) is a catastrophic childhood seizure disorder. It is characterized by extensor and/or flexor spasms, cognitive deterioration and a characteristic EEG pattern of a spike-wave followed by an electrodecremental event (EDR), which refers to the flattening of the EEG waveform. The mechanism/circuitry contributing to the disorder is unknown. Children with Down Syndrome (DS) are especially vulnerable to IS. In this regard, we chose to use a mouse model of DS, the Ts65Dn mutant mouse (Ts) to create an animal model of IS in DS (Cortez et al. 2009). This model entails the treatment of Ts mice with a GABA(B)R agonist to which these mutant animals are equisitely sensitive. Typically, we use γ- butyrolactone [GBL], the prodrug of γ- hydroxybutyric acid [GHB] in this regard. GBL produces a phenotype in the Ts mice that recapitulates the semiological, developmental, and electrophysiological features of IS in children with DS. One of the genes triplicated in Ts mice is the *kcnj6* gene which codes for the GIRK2 protein. We have demonstrated that when we carried out a genetic and pharmacological knockdown of GIRK2 in Ts to create a Ts mouse that was disomic for GIRK2 (Ts65Dn-GIRK2$^{+/+/-}$), we rescued the spasms and EDR to WT levels in the resultant Ts65Dn- GIRK2$^{+/+/-}$ (Joshi et al. 2016). These data indicate that the excess GIRK2 is necessary to confer susceptibility to IS in a DS mouse model. Recently, we asked the question whether the excess GIRK2 is sufficient to confer susceptibility to IS in a DS mouse model. To address this question, we used *kcnj6* triploid mice to determine whether excess GIRK2 alone is sufficient to confer susceptibility to IS. We now show that *kcnj6* triploid mice did not show increased spasms or

EDR compared to WT mice. Therefore, excess GIRK2 is necessary, but not sufficient to confer susceptibility to IS in DS. It is therefore likely that GIRK2 is working in concert with another factor or factors in Ts that produce the IS phenotype. This study is important, because it helps us elucidate the mechanism of IS in DS, thereby providing potential therapeutic interventions in the future.

Disclosures: **K. Joshi:** None. **L. Shen:** None. **M.A. Cortez:** None. **O. Snead:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.07/Q8

Topic: B.11. Epilepsy

Title: Model-guided neurostimulation reduces epileptiform activity in mice

Authors: *J. MODOLO, F. MINA, P. BENQUET, F. WENDLING
INSERM U1099, Rennes, France

Abstract: Drug-refractory epileptic patients have little therapeutic options, since surgical resection is an option for only 10-15% of these patients. Neurostimulation therapy therefore holds great promises for managing these patients. However, one major roadblock is that the involved mechanisms are not fully elucidated, and rational identification of optimal stimulation parameters is still an open issue.
To overcome this roadblock, we developed a model-guided approach consisting in using a biologically grounded neural mass model of brain tissue to identify *in silico* potential candidates for neuromodulation therapy. The model reproduced successfully hippocampal paroxysmal discharges (HPD) characteristics recorded experimentally. Using this model, we explored a simple DC stimulation protocol, and identified an optimal stimulation polarity along with a candidate stimulation site, which should result in a decrease of hyperexcitability.
We tested model predictions *in vivo* using a kainate mouse model of epilepsy. Hyperexcitability was quantified using the number and duration of HPD, which are sustained (up to 60 seconds) epileptiform discharges in the epileptic hippocampus originating in the dentate gyrus (DG). We stimulated the DG in a group of n=5 kainate mice using 11 blocks of 50 seconds of cathodal stimulation (1 µA intensity, electrodes 400 µm apart, 125 µm diameter). Our results revealed a significant decrease in HPD duration and occurrence rate, confirming *in silico* model predictions. Overall, we successfully identified a neurostimulation protocol that decreases hyperexcitability in epileptic mice, based on a model-guided approach. The next step of this model-guided strategy will be to develop neurostimulation protocols based on biphasic, charge-balanced stimulation waveforms usable safely in clinical applications.

Disclosures: **J. Modolo:** None. **F. Mina:** None. **P. Benquet:** None. **F. Wendling:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.08/Q9

Topic: B.11. Epilepsy

Support: CMSRU Research Fund

Title: Preliminary characterization of a new genetic mouse model of adult-onset epilepsy

Authors: *T. N. FERRARO[1], L. YOUNG[1], R. DACI[1], I. SALEM[2], A. BATTERMAN[1], D. MILLER[3], F. PARDO-MANUEL DE VILLENA[3], R. J. BUONO[1], L. D. SIRACUSA[2]
[1]Biomed. Sci., Cooper Med. Sch. of Rowan Univ., Camden, NJ; [2]Microbiology and Immunol., Thomas Jefferson Univ., Philadelphia, PA; [3]Genet., Univ. of North Carolina, Chapel Hill, NC

Abstract: Epilepsy is a common neurological disease with highest incidence in the youngest and oldest segments of the population. Genetic mouse models have led to important clues regarding the nature of epilepsy; however, the phenotypes of known mouse mutants that express spontaneous, recurring seizures emerge early in life, generally by one month of age. We report here the preliminary characterization of a novel mouse model of epilepsy in which generalized seizures are observed in adult animals, not before 5 months of age, and as late as 18 months of age. The index case was a male mouse identified in a cohort from the Collaborative Cross strain CC039/Unc. This male successfully mated, and the seizure phenotype has now been propagated through 4 generations, with more than one-third of offspring (7 of 18) exhibiting spontaneous seizures, including both males and females. There is a significantly longer latency to the onset of seizures in females compared to males. Whole-genome single nucleotide polymorphism analysis of DNA from a number of seizure and non-seizure CC039/Unc mice identified several genomic regions of heterozygosity that are being assessed with regard to their segregation with phenotype. We also discovered a 350 kilobase deletion on chromosome 16 which may affect the promotor sequence of a novel candidate gene, and this factor too is being evaluated in association with phenotype. In further studies, a CC039/Unc male with seizures was mated with females from the FVB/NJ inbred strain. To date, 30 F1 offspring have been generated, 9 of which (4 females; 5 males) have started to express seizures. Another CC039/Unc male with seizures was mated to FVB/NJ females, and their F1 offspring (16 females; 11 males) were tested for electrical seizure threshold at 10-12 months of age. No relationship between seizure threshold and spontaneous seizure phenotype was observed in this cohort. Preliminary characterization of this new mutant mouse strain with spontaneous seizures suggests that the phenotype is inherited in an autosomal dominant manner, but with incomplete penetrance. Another possibility is that the model reflects a two-locus genetic mechanism. An N2 backcross mapping strategy is being followed to localize

the chromosomal position of the factor(s) mediating this novel seizure phenotype. Genetic dissection of this model may lead to new information relevant to adult-onset epilepsy in humans.

Disclosures: **T.N. Ferraro:** None. **L. Young:** None. **R. Daci:** None. **I. Salem:** None. **A. Batterman:** None. **D. Miller:** None. **F. Pardo-Manuel de Villena:** None. **R.J. Buono:** None. **L.D. Siracusa:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.09/Q10

Topic: B.11. Epilepsy

Support: CIHR Operating Grant

NSERC Discovery Grant

Title: Theta and high-frequency oscillation phase-amplitude coupling in stress-induced seizures following traumatic brain injury

Authors: ***P. S. JUNG**[1,2], C. NARLA[1,2], F. BAUTISTA[1,2], J. C. MARTINEZ-TRUJILLO[1,2], M. O. POULTER[1,2]
[1]Western Univ., London, ON, Canada; [2]Robarts Res. Inst., London, ON, Canada

Abstract: Introduction: Traumatic brain injury (TBI) is a substantial cause of epilepsy, accounting for 30% of all cases. In addition, stress has been shown to increase the incidence and severity of seizures following TBI. However, the mechanism underlying the interaction among TBI, seizures, and stress in epilepsy is unclear. High-frequency oscillations (HFOs), biomarkers of seizure genesis, can be informative in understanding this interaction, especially when considered in relation to simultaneous oscillations in slower frequency bands. Furthermore, corticotropin-releasing factor (CRF) released in the amygdala during stress has been shown to be involved in the exacerbation of seizures. Here, we investigate cross-frequency coupling between theta and HFO bands occurring in the amygdala during stress in a brain-injured rat model, with and without a CRF receptor antagonist.
Hypothesis: CRF receptor antagonist decouples the coupling between theta phase and HFO power present during stress in brain-injured rats.
Methods: We used the controlled cortical impact model to simulate TBI in adult male Sprague-Dawley rats (n=6), tail pinch to effect stress, and a CRF receptor 1 (CRFR1) antagonist to attenuate the stress response. Recordings from electrodes implanted *in vivo* in the injured rat amygdala were taken during tail pinch, then repeated 1 hour after intraperitoneal injection of CRFR1 antagonist, and finally after washout 4 hours post-injection. The strength of coupling

between theta phase and HFO power for each recording was quantified using the Modulation Index (MI; Tort et al. 2010 J Neurophysiol 104: 1195-1210). Results: In TBI but not sham-TBI rats, a tail pinch stressor induced an increase in HFO activity (> 120 Hz) in the basolateral amygdala. We observed that the power of high-frequency oscillations was coupled to the phase of the theta band during stress (mean MI = 0.0193 ± 0.0052). This relationship was decoupled 1 hour after a CRF receptor antagonist injection (mean MI = 0.0044 ± 0.0017), but recoupled 4 hours later after washout (mean MI = 0.0197 ± 0.0077). As well, rats exhibited decreased epileptic behaviour during stress when the CRFR1 antagonist was present, compared to baseline and washout.
Conclusion: The power of HFOs was found to be coupled to the phase of simultaneous theta oscillations during stress in the TBI-affected rat. Furthermore, this phase-amplitude coupling decoupled when CRFR1 antagonist was present, indicating the role of stress in this response. Our findings provide insight into the mechanism by which stress and epilepsy interact in individuals affected by TBI.

Disclosures: **P.S. Jung:** None. **C. Narla:** None. **F. Bautista:** None. **J.C. Martinez-Trujillo:** None. **M.O. Poulter:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.10/Q11

Topic: B.11. Epilepsy

Support: CNPq (Science without Borders)

Title: Exacerbated epileptic phenotype in miR-22 knockout mice

Authors: *L. F. ALMEIDA SILVA, C. R. RESCHKE, T. ENGEL, D. C. HENSHALL
Physiol. and Med. Physics, Royal Col. of Surgeons In Ireland, Dublin, Ireland

Abstract: Epilepsy is a brain disease that affects over 50 million people worldwide and is characterized by recurrent seizures. Temporal lobe epilepsy is the most common form of epilepsy in adults and these patients present high pharmacoresistance. Acquired epilepsy is associated with large-scale changes in gene expression which underlie the cell and network-level changes during epileptogenesis. Evidence emerged that miRNAs, a class of non-coding RNAs that fine tune gene expression, play an important role in epilepsy. MiR-22 regulates key epileptogenic-related processes, such as inflammation and apoptosis. Our group detected miR-22 as the most expressed miRNA in the contralateral hippocampus after status epilepticus induced by kainic acid intra-amygdala injection. In this model, the contralateral hippocampus experiences seizures, however does not present neuronal damage. Thus, the contralateral hippocampus could

serve as a source of anti-epileptic molecules. Moreover, we found that silencing miR-22 exacerbates epilepsy. Here, we sought genetic evidence that miR-22 protects against epilepsy. Firstly, we characterized naïve miR-22$^{-/-}$ mice and compared to wild-type littermates. No differences were observed in the brain morphology, gliosis or astrocytosis. We also analyzed levels of miRNAs that are relevant in epilepsy and epileptogenic process. No difference was observed in the level of these miRNAs. When injected with KA intra-amygdala mice lacking miR-22 presented similar seizure severity, measured by EEG total power and amplitude, when compared to wild-type mice. This EEG phenotype was followed by similar neuronal damage in the hippocampus. Interestingly, miR-22 knockout mice presented increased number of spontaneous seizures, when monitored for 2 weeks (average of 19 seizures/day for miR-22$^{-/-}$, and 2 seizures/ day for miR-22$^{+/+}$). Here, we also verified that the intra-nasal administration of miR-22 mimics can be an effective delivery route, once we were able to increase miR-22 hippocampal levels at 4h after administration. These genetic and pharmacological data indicate that miR-22 plays a role in the pathophysiology of epilepsy, and that targeting this miRNA may be a promising approach for epilepsy treatment.

Disclosures: L.F. Almeida Silva: None. **C.R. Reschke:** None. **T. Engel:** None. **D.C. Henshall:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.11/Q12

Topic: B.11. Epilepsy

Title: Role of dorsal medial hypothalamus (DMH) in phenotype of spasms

Authors: *C.-R. CHERN, L. VELISEK
Dept. of Cell Biol. and Anat., New York Med. Col., Valhalla, NY

Abstract: Our two-hit model of infantile spasms consists of prenatal priming with betamethasone and postnatal trigger of spasms with N-methyl-D-aspartic acid (NMDA) in infant rats and involves activation of hypothalamus. Additionally, in our previous study microinfusions of a $GABA_A$ receptor agonist muscimol into the paraventricular hypothalamic nucleus (PVN) *per se* (without any additional trigger) induced a specific phenotype of spasms in rat pups. If combined with a trigger of spasms utilizing NMDA, this muscimol-induced phenotype of spasms appears before NMDA becomes effective. Dorsal medial hypothalamus (DMH) is one of the hypothalamic nuclei projecting directly to PVN. In this project we determined the role of DMH in the control of spasms by functional inhibition of DMH with muscimol microinfusions. We found that muscimol microinfused into DMH modifies the NMDA-induced phenotype of spasms compared to vehicle controls. In muscimol infused animals, we observed an early onset of loss of

posture with imposed long lasting clonic seizures, which was a phenotype that typically occurs during a terminal stage of the NMDA-induced spasms without muscimol microinfusion. Initially, these clonic seizures are interrupted with occasional brief spasms but then advance into long lasting spasms. Additionally, muscimol extended period of spasm occurrence: After NMDA trigger in saline-infused pups, the spasms terminate within 60-75 minutes. In contrast, in muscimol-infused animals, these long-lasting spasms can persist for at least four hours. Our finding suggests that inhibition of DMH activity makes the phenotype of spasms significantly more severe suggesting attenuating role of DMH in this seizure syndrome.

Disclosures: C. Chern: None. **L. Velisek:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.12/R1

Topic: B.11. Epilepsy

Support: NIH NS065877

NIH NS033310

Title: Decrease of gamma event coupling in hippocampal-prefrontal cortex associated with epileptogenesis

Authors: *L. LI[1,2], J. ALMAJANO[1], J. ENGEL[1,2,3], A. BRAGIN[1,2]
[1]Neurol., [2]Brain Res. Inst., [3]Neurobio. and Psychiatry and Biobehavioral Sci., Univ. of California Los Angeles, Los Angeles, CA

Abstract: It is accepted that epilepsy is a network phenomenon with changes of both functional and anatomical brain networks. The medial prefrontal cortex (PFC) and hippocampus (HIP) are key brain areas for memory consolidation processes. We hypothesize that dysfunction of these networks could lead to memory impairment and could be early biomarkers of epileptogenesis. Our study aims to investigate PFC and HIP network changes during the latent period in an animal model of temporal lobe epilepsy (TLE). Experiments were conducted on 15 rats with 8 microelectrodes implanted in symmetrical sites of PFC and HIP. After one week of baseline recordings KA was injected in the left anterior hippocampus to evoke status epilepticus (SE). Animals were divided into two groups: with epilepsy and without epilepsy during/after 2 month of recording. Functional connectivity was quantified by the measurement of *gamma event coupling* (GFC), in which Shannon entropy was computed. A sequence of gamma events with amplitude higher than 3SD were considered as *Gamma oscillations (GO)*. Peri-event histogram analysis was performed to study the synchrony between GEC and GO and multi-unit discharges

(MUDs) within HIP and between HIP-PFC. Group-level comparisons of GEC and synchrony of GS and MUDs were performed between latent period and baseline. We found a decrease of GEC between HIP & PFC ipsilateral to the KA lesion during the 1st week after SE in rats that later developed epilepsy, while no significant changes in GEC between HIP and other recorded brain areas. We further determined that this decrease in GES was associated with decreased of synchrony of MUD in both in HIP and PFC on the lesion side. No significant change of GEC and synchrony in MUD between HIP and PFC in the non-epilepsy group. Furthermore, we found that 40% of GEC occurred during PFC gamma oscillations (PFC-GO) in all animals, although the total number of PFC-GO was much less in the group with epilepsy. Our results also revealed a significant decrease in synchrony of HIP MUD during PFC-GO on the lesion side in the group with epilepsy, but no changes were observed in the group without epilepsy. The changes in functional connectivity related to epileptogenesis are detectable using GEC, while GO is less sensitive in the assessment of epileptogenic related connectivity changes. Because gamma activity mostly is related with activity of interneuronal networks, changes in GEC may indicate deterioration in the relationship between HIP and PFC via inhibitory mechanisms. Interruption of this network could be an early biomarker of epileptogenesis and may have implication for the detecting of psychiatric comorbidities during epileptogenesis.

Disclosures: L. Li: None. **J. Almajano:** None. **J. Engel:** None. **A. Bragin:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.13/R2

Topic: B.11. Epilepsy

Support: MRC Grant MR/J013250/1

Title: Characteristics of evolving epileptiform activity in different cortical areas in brain slices

Authors: *N. CODADU[1], A. TREVELYAN[2]
[2]Inst. of Neurosci., [1]Newcastle Univ., Newcastle Upon Tyne, United Kingdom

Abstract: Understanding regional differences in the susceptibility to the development of epileptic activity and the nature of brain state transitions in its evolution remains a major goal for epilepsy research. One such transition, of potentially great importance for drug development, is from a pharmaco-sensitive to a pharmaco-resistant pattern of discharges that occurs in a widely used, rat, in vitro model (low Mg^{2+} bathing medium). Early epileptiform activity, including interictal discharges and sustained events that mimic clinical tonic-clonic seizure activity, is suppressed by many anti-epileptic agents (Dreier et al., 1998). There then follows a transition into a late pattern of repeated discharges that are resistant to the same drugs. The nature of this

critical transition, however, has remained a mystery. We have replicated these patterns in mouse brain slices, and report here that the transition reflects a change in the involvement of hippocampal networks and how they interact with the other cortical networks. Early ictal discharges only involve neocortical and entorhinal networks. Epileptiform events in hippocampus develop much later, but once these start, they quickly entrain the neocortex to their, more regular, discharge pattern. Notably, this entrainment persists following transection of the major axonal pathways between hippocampus and neocortex, indicating that it is mediated through a non-synaptic route. Analyses of the timing of these events demonstrate that the hippocampal activity is the pacemaker of this late-stage pattern of discharges. In a second in vitro model of epilepsy, induced instead by bathing brain slices in the K^+ channel blocker 4-aminopyridine (4AP), activity evolves over a period of tens of minutes to show both tonic-clonic like discharges, and eventually, a pattern of repeated spike and wave discharges. In these regards, both low Mg^{2+} and 4AP models appear similar, but the evolution to this stage is very different. While the low Mg^{2+} model involves hippocampal networks only in the late stage, the 4AP model induces pathological discharges in the hippocampal networks very early. Our results illustrate the multifaceted nature of epileptiform activity, providing many metrics for characterising drug actions, and for understanding phenotypes in genetic models of epilepsy.

Disclosures: **N. Codadu:** None. **A. Trevelyan:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.14/R3

Topic: B.11. Epilepsy

Support: Swedish Research Council

The Royal Physiographic Society

EPITARGET (EU grant)

Title: Transhemispheric network interaction for hippocampal ictogenesis revealed by combined optogenetic and chemogenetic interrogation *In vivo*

Authors: *F. BERGLIND, M. S. ANDERSSON, M. KOKAIA
Epilepsy Center, Lund Univ., Lund, Sweden

Abstract: The hallmark of epilepsy is synchronous repetitive firing of neuronal ensembles, manifested as seizures. The manner in which neurons are recruited to this network in the initial stages of disease development and the minimal neuronal circuit needed to generate progressive seizure activity is, however, not known. By selectively activating principal neurons using a

protocol for repetitive optogenetic stimulation in one hemisphere of the hippocampus of anaesthetized transgenic CaMKII-ChR2 mice, we observed a rapid development (within 15-25 stimulations, 1-2 hours) of progressively intensifying epileptic afterdischarges (ADs) generated concurrently in both hemispheres, as measured by bilateral local field potential (LFP) recordings. This corroborated earlier data suggesting that progressively intensifying ADs strongly correlated with activation of the contralateral dentate gyrus (DG) area of the hippocampus. To further test this hypothesis we utilized a loss-of-function approach: when the DG contralateral to optogenetic stimulation was inhibited by Gi-DREADD (a designer receptor exclusively activated by the otherwise biologically inert molecule CNO), the progressive intensification of ADs was abrogated. These findings suggest that transhemispheric bilateral DG activation is required for the establishment of the minimal hyperexcitable neuronal circuit that supports early stage progressive intensification of ADs in the hippocampus, which may eventually lead to permanent changes in the hippocampal networks towards increased excitability and development of epilepsy.

Disclosures: F. Berglind: None. **M.S. Andersson:** None. **M. Kokaia:** None.

Poster

383. Epilepsy: EEG signatures and Animal models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 383.15/R4

Topic: B.11. Epilepsy

Title: Bilateral evolution of fast-ripples and interictal spikes during epileptogenesis

Authors: *P. BENQUET[1,2], J. MODOLO[3], M. VERIN[3], F. WENDLING[3]
[1]INSERM U1099 -LTSI, Rennes, France; [2]Univ. of Rennes 1, Rennes, France; [3]INSERM U1099-LTSI, Rennes, France

Abstract: In focal epilepsies, seizures are triggered in a circumscribed brain region often referred to as the epileptogenic zone (EZ). Accurate EZ delineation is an essential step in planning the best therapeutic strategy. To achieve this, intracranial electrophysiological recordings remain the most precise identification method to record dysfunctional biomarkers characteristic of underlying epileptogenic networks.
Several types of interictal activity are observed in depth-EEG signals recorded with intracerebral electrodes in the epileptic brain, such as fast-ripples (FRs), which are brief (a few tens of milliseconds), high-frequency oscillations (HFOs; 250-600 Hz). FRs have been a topic of increasing interest for the past two decades since i) FRs seems to be generated locally within the EZ; and ii) resection of brain tissue producing FRs demonstrated the potentially significant diagnostic value of HFOs in focal epilepsies. To a lesser extent, interictal epileptic spikes (IESs) have also been considered as potentially useful makers, however one drawback is that this

biomarker can propagate outside the EZ. The generation and diffusion of these biomarkers during the slow pathological plasticity associated with the epileptogenesis phase is still not well characterized.
In this study, we used the *in vivo* kaïnate mouse model of epilepsy to study the evolution, characteristics and propagation of fast-ripples and epileptic spikes during epileptogenesis both in the EZ (kaïnate injected side, right hippocampus) but also in the control hippocampus (non-injected side, left hippocampus).
During the three weeks of epileptogenesis, the occurrence rate of IESs and FRs not only increased, but a significant difference in term of localization and propagation of these biomarkers was quantified. Significant changes in IES morphology and in FRs spectral dynamic were also recorded. Bilateral recordings revealed that IES generated in the left hippocampus could trigger FR in the contralateral side. We used computational modeling to provide mechanistic insights regarding the evolution of these biomarkers.

Disclosures: **P. Benquet:** None. **J. Modolo:** None. **M. Verin:** None. **F. Wendling:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.01/R5

Topic: B.12. Glial Mechanisms

Support: State University of New York

State University of New York Research Foundation

Title: Astrocyte remodeling alters short- and long-term excitatory synaptic transmission in the mouse hippocampus

Authors: ***J. MCCAULEY**[1], A. M. SWEENEY[2], K. E. FLEMING[3], M. F. RODRIGUEZ[5], E. T. MARTIN[4], A. A. SOUSA[6], R. D. LEAPMAN[7], A. SCIMEMI[8]
[1]Biol., Univ. At Albany State Univ. of New York, Albany, NY; [2]Univ. at Rochester, Rochester, NY; [3]Biol., [4]Univ. at Albany, SUNY, Albany, NY; [5]SUNY Oneonta, Oneonta, NY; [6]Univ. of San Paulo, San Paulo, Brazil; [7]NIH, Bethesda, MD; [8]Biol., Univ. at Albany State Univ. of New York, Albany, NY

Abstract: Astrocytes express a variety of G-protein coupled receptors. Type 1 Protease Activated Receptors (PAR1) are G-protein coupled receptors activated by bloodstream serine proteases, like thrombin and plasmin. These proteins diffuse out of the bloodstream under pathological conditions like small hemorrhagic stroke. What is not known is whether PAR1 activation affects glutamate uptake, one of the main function of astrocytes. Here we combine

electrophysiology, imaging and modeling approaches to show that PAR1 activation speeds glutamate clearance by astrocyte transporters in the mouse hippocampus. This effect is not due to changes in glutamate transporter expression or to widespread changes in the diffusion properties of the neuropil. Instead, the faster clearance is associated with rapid restructuring of astrocytic processes in the immediate vicinity of excitatory synapses. This includes changes in the number, surface area and proximity of astrocytic processes to synapses. By using realistic 3D Monte Carlo reaction-diffusion simulations, we show that all these effects can lead to faster glutamate clearance in and out of the synaptic cleft. Astrocyte remodeling due to PAR1 activation alters short- and long-term synaptic transmission. Taken together, these findings identify PAR1 as a key regulator of excitatory synaptic function in the hippocampus and a potential target to prevent cognitive impairment and decline following small hemorrhagic stroke.

Disclosures: J. McCauley: None. **A.M. Sweeney:** None. **K.E. Fleming:** None. **M.F. Rodriguez:** None. **E.T. Martin:** None. **A.A. Sousa:** None. **R.D. Leapman:** None. **A. Scimemi:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.02/R6

Topic: B.12. Glial Mechanisms

Support: NINDS Grant F31NS100259

NINDS Grant R01NS075062

Title: Astrocytes are a primary target for neuronal BDNF: Implications for the regulation of astrocyte morphological complexity

Authors: *L. HOLT[1,3], M. L. OLSEN[2]
[2]Sch. of Neurosci., [1]Virginia Tech., Blacksburg, VA; [3]Cell, Integrative, and Developmental Biol., Univ. of Alabama at Birmingham, Birmingham, AL

Abstract: Most astrocytes are born during the 2-4th postnatal week in rodents. This is followed by a period of morphological maturation including the development and extension of fine dense processes, which eventually enwrap synapses and blood vessels. The developmental time window of astrocyte morphological maturation and refinement is well defined but the molecular mechanisms that underlie this process are not understood. Brain derived neurotrophic factor (BDNF) is a critical growth factor secreted largely by neurons and involved in the development and maturation of neurons, including neuronal growth and synapse refinement. In the current study we demonstrate that astrocytes express high levels of the BDNF receptor TrkB when

compared to neurons. Quantitative PCR indicates astrocytes predominantly express the truncated version of TrkB, TrkB.T1, which lacks the canonical kinase domain. TrkB.T1 expression is highest in astrocytes during the critical period of astrocyte morphological refinement and maturation, a developmental time window that coincides with the highest neuronal BDNF mRNA expression levels. These findings have led us to hypothesize that BDNF/TrkB.T1 signaling is an important mediator of astrocyte morphological maturation. Indeed, preliminary data demonstrates that exposure to BDNF increases astrocyte cellular complexity. Ongoing studies are aimed at understanding the relative importance of BDNF/TrkBT.1 signaling in astrocyte morphological maturation.

Disclosures: L. Holt: None. **M.L. Olsen:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.03/R7

Topic: B.12. Glial Mechanisms

Support: NIH Grant NS089640

Title: Role of NAD^+ availability and Sirt6 expression in the regulation of antioxidant defenses in astrocytes

Authors: *B. HARLAN, K. M. KILLOY, M. PEHAR, M. R. VARGAS
Cell and Mol. Pharmacol., Med. Univ. of South Carolina, Charleston, SC

Abstract: Nicotinamide adenine dinucleotide (NAD^+) is an essential redox molecule and a key player in several signaling pathways that govern fundamental biological processes. In redox reactions, a hydride equivalent is reversibly transferred at the nicotinamide moiety causing a switch between oxidized (NAD^+) and reduced (NADH) forms of the nucleotide. In contrast, NAD^+-dependent signaling involves the cleavage of NAD^+ (degradation) coupled to post-translational modifications of proteins or the production of second messengers. Activation of NAD^+-dependent signaling pathways has a major effect in the capacity of the cell to modulate mitochondrial function and counteract the deleterious effects of increased oxidative stress. Sirtuins (Sir2-like enzymes) are NAD^+-dependent deacylases that play a key role in transcription, DNA repair, metabolism, and oxidative stress resistance. There are seven mammalian sirtuins (Sirt1-7) with diverse subcellular localization, enzymatic activity and protein substrates. Sirt6 is a chromatin-associated nuclear protein involved in DNA repair, telomere maintenance, gene expression, and metabolism. Here we explore the role of Sirt6 over-expression and increased NAD^+ availability in astrocyte antioxidant defenses and astrocyte-motor neuron interaction. Astrocytes play a key role determining the progression of the

neurodegenerative process in amyotrophic lateral sclerosis (ALS). Accordingly, astrocytes over-expressing ALS-linked mutant hSOD1 induce the death of co-cultured motor neurons. We have previously shown that up-regulation of antioxidant defenses reverts astrocyte-mediated neurotoxicity. We observed that increasing NAD^+ availability or Sirt6 over-expression induced Nrf2-driven gene expression and revert the neurotoxic phenotype of $hSOD1^{G93A}$-expressing astrocytes. Together, our results suggest that Sirt6 could be a potential therapeutic target in ALS.

Disclosures: B. Harlan: None. **K.M. Killoy:** None. **M. Pehar:** None. **M.R. Vargas:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.04/R8

Topic: B.12. Glial Mechanisms

Support: NIH Grant 7K01MH097957-02

NIH Grant 1R01NS096100-01

NIH Grant 1R21MH110724-01

Title: Downregulation of Sonic hedgehog in reactive astrocytes after injury

Authors: *R. ALLAHYARI, P. SAKTHIVEL, K. L. CLARK, A. D. R. GARCIA
Biol., Drexel Univ., Philadelphia, PA

Abstract: Following injury to the central nervous system, the molecular signaling pathway Sonic hedgehog (Shh) has been shown to promote various cellular activities, including proliferation of adult neural stem cells (NSCs) and increased neurite outgrowth. Both Shh and its target gene, Gli1, have been shown to be upregulated following injury. This suggests that Shh signaling may play a role in neural repair processes. More specifically, Shh is a known mitogen in the developing nervous system and adult NSC niches, suggesting it may be responsible for regulating injury induced proliferation. We previously identified a subpopulation of astrocytes in the adult cortex that express Gli1. The precise role of Gli1 astrocytes in CNS injury is not well understood. In this study we used molecular genetic strategies to examine Gli1 astrocytes after forebrain stab injury. Our data show that following a forebrain stab injury, there are fewer Gli1 astrocytes in the lesion area at 24 hours, compared to uninjured controls. This reduction in Gli1 cells persists for up to 7 days after injury. Using genetic inducible recombination strategies, our data show that Gli1 astrocytes are not dead, but rather, that Gli1 expression is downregulated. This suggests that Shh signaling is downregulated in response to acute forebrain stab injury. Our data also show that Shh and Gli1 expression returns to the injured cortex 14 days post injury, as confirmed by multiple transgenic mouse models and ddPCR, suggesting that Gli1 astrocytes

undergoing reactive astrogliosis transiently downregulate Shh signaling. These data are consistent with a recent study in the lung epithelium showing that Gli1 is similarly downregulated in mesenchymal progenitor cells following injury. Ongoing studies are aimed at investigating the functional consequences of downregulation of this pathway in the injury response.

Disclosures: **R. Allahyari:** None. **P. Sakthivel:** None. **K.L. Clark:** None. **A.D.R. Garcia:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.05/R9

Topic: B.12. Glial Mechanisms

Support: JSPS KAKENHI (16H01329)

JSPS KAKENHI (17H03988)

JSPS KAKENHI (17H05738)

Title: Optogenetic regulation of cyclic AMP in brain cells *In vivo*

Authors: ***Z. ZHOU**[1], J. ONODERA[1], T. HIRAGI[1], M. ANDOH[1], K. F. TANAKA[2], R. KOYAMA[1], Y. IKEGAYA[1]
[1]Grad Sch. Pharma Sci, Univ. Tokyo, Tokyo, Japan; [2]Keio Univ. Sch. of Med. Dept. of Neuropsychiatry, Tokyo, Japan

Abstract: In most of the optogenetic approaches, microbial opsins are expressed in neurons to elicit or inhibit neuronal activity via light-induced generation of ionic transmembrane currents, leaving other intracellular signalings remain to be optogenetically studied. Here we optogenecically modulated the intracellular levels of cAMP in vivo, using photoactivated adenylyl cyclase (PAC). PAC, which was originally identified as a photosensor of the flagellate *Euglena gracilis*, is activated by blue light and rapidly changes its conformation to synthesize cAMP from ATP. Recently, several new PACs including bPAC from the soil bacterium Beggiatoa have been discovered. We successfully developed a mouse line which carries both bPAC and GFP genes downstream of TetO operator sequences. By crossing these mice with lines that express tTA under different promoters, we managed to express PAC in astrocytes, microglia and neurons, separately. PAC is expressed in most of astrocytes across the brain in Mlc1-PAC mice, almost half of microglia in Iba1-PAC mice, and neurons in specific regions including CA1, habenula, and stiatum and orbital frontal cortex (5HT5b-PAC mice). We confirmed an increase of the intracellular cAMP levels in response to blue light illumination in

cultured astrocytes prepared from Mlc1-PAC mice, using a newly developed red cAMP sensor. Further, in vivo light stimultion in Mlc1-PAC mice induced CREB phosphorylation in PAC-expressing astrocytes and learnig and memory-related behavioral changes. Thus, our TetO-PAC mice will be useful for studying the role of cAMP in brain cells in vivo.

Disclosures: Z. Zhou: None. **J. Onodera:** None. **T. Hiragi:** None. **M. Andoh:** None. **K.F. Tanaka:** None. **R. Koyama:** None. **Y. Ikegaya:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.06/R10

Topic: B.12. Glial Mechanisms

Title: Nanostructered silicon nanowire and electrospun nonofibre polymer to promote astrocytes differentiation and recording *In vitro*

Authors: *E. SARACINO[1], A. I. BORRACHERO-CONEJO[2], V. CIRILLO[3], V. GUARINO[3], A. CONVERTINO[4], L. MAIOLO[4], M. MARRESE[5], M. CAPRINI[6], A. BORRIELLO[3], L. AMBROSIO[3], G. FORTUNATO[4], R. ZAMBONI[7], V. BENFENATI[7]
[1]Inst. per la sintesi organica e la fotoreattività ISOF, Consiglio Nazionale Delle Ricerche Area Della Rice, Bologna, Italy; [2]CNR-Istituto per lo studio dei materiali nanostrutturati ISMN, Bologna, Italy; [3]CNR-Istituto dei Polimeri, Composti e Biomateriali IPCB, Napoli, Italy; [4]CNR-Inst. per elettronica e magnetismo IMEM, Roma, Italy; [5]CNR- Inst. dei Polimeri, Composti e Biomateriali IPCB, Napoli, Italy; [6]Dept. di Biotecnologie e Farmacia, Univ. di Bologna, Bologna, Italy; [7]CNR-Istituto per la sintesi organica e la fotoreattività ISOF, Bologna, Italy

Abstract: Consolidated evidence highlight the importance of astrocytes ion channels and aquaporin 4 (AQP4) in brain physiology and pathological conditions. Studying ion channels and AQP4 in a controlled and reliable in vitro model might promote increasing knowledge on nervous system function and dysfunction or degeneration and on neuro regenerative medicine. However, when astrocytes are grown in culture, they lose their characteristic star-like shape. Also, astrocytes in vitro loose the peculiar polarized localization of AQP4 and ion channels in the plasma membrane observed in astrocytes endfeet *in vivo*. In this view, our goal is to define and characterize cell culture model of astrocytes in vitro resembling the features of astrocytes in vivo and to identify molecular, biophysical and chemical factors that might be relevant in the regulation/modulation of the expression and functionality of ion channels and AQP4 in astrocytic membrane. To address this issue we sought to investigate on the effect on primary rat cortical astrocytes growth and physiology of innovative nanostractured material interface based on inorganic silicon/gold nanowires (SiAunw) and on organic polymers polycaprolactone (PCL) electrospun nanofibers. By means of cell viability assays we found that siAunw as well as PCL

electrospun nanofibers promote strong adhesion of astrocytes without need of additional coating with values comparable to those obtained by using glass coverslips treated with Poly-D-lisine. Optical and fluorescent imaging at different time points revealed that nanostructures promote modification of astrocytes morphology. Scanning Electron Microscopy and Atomic Force Microscopy confirms the capability of astrocytes to respond to substrate topography. We show how interfaces can promote astrocytes differentiation with three dimensional growth and by promoting endfeet sprouting from the cell body lining and enveloping the interface nanostructure. The differentiation is not due to gliotic reaction as GFAP values are unchanged respect to the control. Also, PCL nanofibre alignment promote astrocytes growth and orientation along the fibre with induction of F-actin fibre alignment and vinculin polarization, as revealed by immunofluorescence and fluorescent confocal microscopy. Finally, by whole-cell recording patch-clamp, the eletrophysiological properties of the cells have been characterized, revealing promising features in the expression of ion conductance resembling those expressed by astrocytes in vivo also in the case of PCL fibres with polyaniline nanowires. This work is supported by AFOSR projects ASTROMAT, FA9550 16 1 0502 and ASTRONIR, FA9550-17-1-0502.

Disclosures: **E. Saracino:** None. **A.I. Borrachero-Conejo:** None. **V. Cirillo:** None. **V. Guarino:** None. **A. Convertino:** None. **L. Maiolo:** None. **M. Marrese:** None. **M. Caprini:** None. **A. Borriello:** None. **L. Ambrosio:** None. **G. Fortunato:** None. **R. Zamboni:** None. **V. Benfenati:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.07/S1

Topic: B.12. Glial Mechanisms

Support: Samsung Science & Technology Foundation Grant SSTF-BA1502-13

the ministry of education, science and technology as global frontier project (CISS-2012M3A6A6054204)

Title: Effect of Zinc ions on astrocytic calcium oscillations in mice

Authors: *Y. A. CHO[1], S. KIM[2], S. HWANG[2], K. NOH[3], W.-H. CHO[3], E. BARCELON[3], S. YOON[3], S. LEE[3], S. JUN[2]
[1]Electronics Engin., [2]Electronic & Electrical Engin., Ewha Womans Univ., Seoul-City, Korea, Republic of; [3]Sch. of Dent., Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: Communication between neuron and astrocyte has become emerging issue in brain science. Recently intracellular calcium oscillations in astrocytes are spotlighted for encoding memory functions. It was known that presynaptic glutamate release increases the intracellular calcium level of neighboring astrocyte and presumably induces oscillation of astrocytic intracellular calcium throughout the brain. In addition, it was reported that labile zinc ions affect the signaling of neurons and astrocytes at specific brain regions such as hippocampus. Zinc ions exist in glutamatergic synapses and believed to regulate the glutamate release in order to maintain the glutamate homeostasis and to prevent glutamate excitotoxicity. This study investigates the zinc influence in astrocytic Ca^{2+} oscillation with optical imaging technique. Hippocampal brain slice from astrocyte-specific GCaMP5 transgenic mice was prepared for calcium imaging to monitor the intracellular calcium level of astrocyte. Since the activation of astrocytes are closely related with neurons, neuronal activities was electrically recorded via microelectrode arrays simultaneously with calcium imaging. In order to trigger Ca^{2+} oscillation in astrocytes, neurons were electrically or chemically evoked. When external zinc ions were applied to the brain slices, the frequency change of astrocytic calcium oscillation was observed. This study showed the possibility that zinc ions modulated the astrocytic Ca^{2+} oscillation in the brain.

Disclosures: Y.A. Cho: None. **S. Kim:** None. **S. Hwang:** None. **K. Noh:** None. **W. Cho:** None. **E. Barcelon:** None. **S. Yoon:** None. **S. Lee:** None. **S. Jun:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.08/S2

Topic: B.12. Glial Mechanisms

Support: NIH Grant R01MH110504

Title: Automated analysis of in-vivo astrocyte activities from large-scale calcium imaging data

Authors: *G. YU[1], Y. WANG[1], K. POSKANZER[2]
[1]Virginia Tech., Arlington, VA; [2]Biochem. and Biophysics, Univ. of California, San Francisco, San Francisco, CA

Abstract: The advent of modern microscopy and highly sensitive protein calcium sensors enables the recording of astrocyte activities on large population of cells over a long time period. However, sophisticated analytical tools lag far behind the impressive capability of generating the data. The analysis of astrocyte activities is particularly challenging due to the complexity of astrocyte calcium signaling. The current practice of using such information-rich data relies heavily on manual analysis, which not only limits the scale of study but also potentially

introduces analysis bias and misses subtle yet important information. Previously, we built an automated data-driven and robust tool to analyze in-vitro astrocyte data. Here, applying advanced machine learning theory and using computational techniques, we extend the tool to deal with in-vivo astrocyte data. The in-vivo data has special characteristics that need to be carefully modeled. Firstly, we allow for flexible signal propagation. For different pixels associated with the same calcium event, propagation patterns cannot be accounted for by time shift only, instead that the signal can shrink, expand or be initiated asynchronously but terminated simultaneously. Propagation patterns are also likely to differ from event to event. Secondly, an event-oriented view is taken. In in-vitro data, we have assumed all pixels in an ROI (region of interest) share a common activity pattern (curve). This is not true for in-vivo data, because some pixels may participate one event but keep silent for another event. Essentially, the concept of ROI is inappropriate due to the fact that the given region is heterogeneous with respect to different events. Hence, we take an event-oriented view, with an event defined jointly by its spatial occupancy and temporal dynamics. Lastly, but not least importantly, computational techniques and algorithmic innovation are exercised to speed up the analysis. The in-vivo recording can easily result in a large data set. For example, we routinely see data set with file size over gigabytes and with hundreds of thousands calcium events.

Disclosures: **G. Yu:** None. **Y. Wang:** None. **K. Poskanzer:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.09/S3

Topic: B.12. Glial Mechanisms

Support: T32-DA7288

R015369

Title: Self administration of cocaine or heroin reduces S-glutathionylation of cofilin and enhances glial fine process motility

Authors: *A. KRUYER, M. SCOFIELD, J. UYS, P. W. KALIVAS
Med. Univ. of South Carolina, Charleston, SC

Abstract: Spillover of synaptically-released glutamate in the nucleus accumbens (NA) is a fundamental feature underlying relapse to addictive drugs. Perisynaptic astroglial processes containing glutamate transporters, which maintain glutamatergic tone at NA synapses, retract during extinction from cocaine or heroin use, though the cellular mediators of this process are not understood. We hypothesize that repeated acute oxidative stress produced in the NA by

cocaine or heroin self-administration leads to long-lasting post-translational de-glutathionylation of the actin-binding protein cofilin, an important regulator of glial fine process morphology. Using a variation of the *in situ* S-glutathionylation switch protocol, we have examined the S-glutathionylation of cofilin *ex vivo* in the context of extinction from cocaine or heroin self-administration and reinstated drug seeking. Following at least 10 days of extinction from cocaine or heroin self-administration we found an enduring reduction in the glutathionylation state of cofilin. Furthermore, when cocaine-trained animals underwent cue-induced reinstatement of cocaine seeking, S-glutathionylation of cofilin was also decreased, and the decrease was strongly correlated with the number of active lever presses enacted during the 15-minute reinstatement session prior to extraction of NA tissue. Cofilin is a well-known regulator of actin and cell morphology, and may contribute to drug-induced changes in the presence of fine glial processes in the vicinity of excitatory synapses in the NA. Indeed, we find that after extinction from cocaine or heroin self-administration there is a reduction in proximity of astroglial surface to the synaptic marker Synapsin I, and that after 15 minutes of cue-induced heroin seeking, astroglial processes are in greater proximity to the synaptic marker. Our preliminary data support the overarching hypothesis that post-translational modification based on tissue redox status is a mechanism by which drugs of abuse may encode long-lasting changes in glutamatergic circuitry.

Disclosures: **A. Kruyer:** None. **M. Scofield:** None. **J. Uys:** None. **P.W. Kalivas:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.10/S4

Topic: B.12. Glial Mechanisms

Support: NRF-2011-0021866the Korea Healthcare Technology R&D Project

the National Research Foundation of Korea HI3C1451

the Brain Korea 21 PLUS program

Title: Hippocampus-based contextual memory alters the morphological characteristics of astrocytes in the dentate gyrus

Authors: *M. CHOI
Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: Astrocytes have been reported to exist in two states, the resting and the reactive states. Morphological changes in the reactive state of astrocytes include an increase in thickness and number of processes, and an increase in the size of the cell body. Molecular changes also occur, such as an increase in the expression of glial fibrillary acidic protein (GFAP). However, the

morphological and molecular changes during the process of learning and memory have not been elucidated. In the current study, we subjected Fvb/n mice to contextual fear conditioning, and checked for morphological and molecular changes in astrocytes. 1 h after fear conditioning, type II and type III astrocytes exhibited a unique status with an increased number of processes and decreased GFAP expression which differed from the typical resting or reactive state. In addition, the protein level of excitatory excitatory amino acid transporter 2 (EAAT2) was increased 1 h to 24 h after contextual fear conditioning while EAAT1 did not show any alterations. Connexin 43 (Cx43) protein was found to be increased at 24 h after fear conditioning. These data suggest that hippocampus-based contextual memory process induces changes in the status of astrocytes towards a novel status different from typical resting or reactive states. These morphological and molecular changes may be in line with functional changes.

Disclosures: M. Choi: None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.11/T1

Topic: B.12. Glial Mechanisms

Support: FRGS 2014-1

Title: Investigating the underlying mechanisms of programmed cell death in human hippocampal astrocyte cell line following exposure to hypoxia

Authors: *J. M. ABDULLAH, Prof[1,2], S. MUTHURAJU[1,2], N. BINTI M.NOR NAZLI[1,2], A. MOHAMED YUSOFF[1,2], F. AHMAD[1,2], M. MUSTAFA[1,2], H. JAAFAR[3], S. SHAMSUDDIN[4]
[1]Ctr. for Neurosci. Services and Res., P3Neuro,Universiti Sains Malaysia, Kota Bharu, Malaysia; [2]Dept. of Neurosciences, Sch. of Med. Sciences, Universiti Sains Malaysia, Kota Bharu, Kelantan, Malaysia; [3]Dept. of Pathology, [4]Sch. of health Sci., Sch. of Med. Sciences, Universiti Sains Malaysia, Kota Bharu, Malaysia

Abstract: Oxygen is the most vital element in maintaining the homeostasis and ensures efficiency of human body systems. Lacking of this chemical element or disruption of balance between its supply and its demand may cause disparity changes. A condition where oxygen availability is limited can be described as hypoxia. Many studies used different kinds of cell lines of animals as well as human to find out the mechanisms of cell death and duration following hypoxia. However, no study has not been evaluated the effect of hypoxia particularly on human hippocampal astrocyte cell line. Therefore, the present study aimed to investigate the underlying mechanisms of cell death in human hippocampal astrocyte cell line. Following exposure to hypoxia. Based on the preliminary screening, almost 80% of cell death occurred after 20 min and

60% cell death occurred in 15 min after exposed to chronic hypoxia, 3% of oxygen level ($p<0.05$). From the data gained, 15 minutes was chosen as the time point and the cells were exposed to different oxygen percentage. Analysis from Trypan blue viability assay showed about 15% of cells were dead in 15% oxygen, 25% dead cells in 10% oxygen, 48% dead cells in 5% oxygen and 65% dead cells in 3% oxygen ($p<0.05$). For the immunofluorescence assay, a reliable marker GFAP was used in order to portray the architecture and morphology of astrocytes cells. Fluorescence scanning microscope revealed a filamentous and clear nucleus appearance in a control. In contrast, the rupture nuclei along with no rigid structure of cell were displayed in chronic hypoxia group, the 3% oxygen exposure. The control and hypoxia cells also were stained with the Annexin V FITC and then observed under a fluorescence microscope. Astrocyte cells after hypoxia showed higher expression of nuclei but not in control. Merged between PI and FITC clearly showed the differences of nuclei expression between the control and hypoxia exposed group. Along with that, the HIF-1 staining was performed to confirm the cell death due to hypoxia exposure. Based on the fluorescence microscope viewed, there were dramatic expression of HIF-1α was displayed in exposed astrocyte cells compare to the control. Besides, GAPDH, HIF-1α and Bcl_2 genes were observed in RT-PCR. Thus, we could conclude that morphological changes of astrocyte cells begins within 15 minutes with chronic hypoxia which clearly showed the damages as their nuclei ruptured and expression of apoptotic genes. **Key words:** Hypoxia, human hippocampal astrocytes, oxygen percentage, cell viability, morphological changes, FITC Annexin V staining, GFAP marker, HIF-1α, GAPDH, Bcl_2.

Disclosures: J.M. Abdullah: None. **S. Muthuraju:** None. **N. Binti M.Nor Nazli:** None. **A. Mohamed Yusoff:** None. **F. Ahmad:** None. **M. Mustafa:** None. **H. Jaafar:** None. **S. Shamsuddin:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.12/T2

Topic: B.12. Glial Mechanisms

Support: MOST 103-2321-B-010-011 from Ministry of Science and Technology, Taiwan

Title: Aryl hydrocarbon receptor mediates feedback regulation of lipopolysaccharide-induced proinflammatory astrogliosis to maintain cognitive functions in mice

Authors: *Y.-L. GAN[1], S.-H. WU[1], C.-H. LIN[2], Y.-J. HUANG[1], F.-S. SHIE[3], H.-C. LIN[1,4], C.-J. JENG[4,5], Y.-H. LEE[1,4]
[1]Inst. of Physiol., Natl. Yang-Ming Univ., Taipei, Taiwan; [2]Dept. of Nursing, Univ. of Kang Ning, Taipei, Taiwan; [3]Ctr. For Neuropsychiatric Research, Natl. Hlth. Res. Inst., Zhunan Town,

Taiwan; [4]Brain Res. Center, Natl. Yang-Ming Univ., Taipei, Taiwan; [5]Inst. of Anat. & Cell Biology, Natl. Yang-Ming University, Taipei, Taiwan, Taipei, Taiwan

Abstract: Inflammatory insult-induced astrogliosis plays important roles in the neuronal survival and injury repair after brain injuries. Previous studies have shown that indoleamine 2, 3-dioxygenase (IDO)-aryl hydrocarbon receptor (AhR) pathway has immune modulatory effect on peripheral immune responses. We previously showed that AhR, a ligand-activated transcription factor activated by environmental toxins and tryptophan metabolites, can aggravate N-methyl-D-aspartate receptors (NMDARs)-mediated excitotoxicity. Peripheral inflammation has been shown to induce neuroinflammation that is associated with mental and cognitive impairment in animal models. In this study, we investigated the role of AhR in astrocyte-neuron interaction under lipopolysaccharides (LPS) stimulation. We found that LPS treatment could activate AhR as well as both AhR and IDO were induced by LPS in cultured astrocytes. Besides, knockdown of AhR profoundly enhanced the LPS-induced IDO expression. Conversely, IDO knockdown decreased LPS-induced AhR expression. As a result, the LPS-induced proinflammatory cytokine IL-6 and TNFα expression were found negatively regulated by the LPS-induced AhR and IDO expression in astrocytes. Notably, the peripheral LPS treatment impaired novelty-related social-cognitive behaviors including novel object recognition and social novelty in WT mice, and both effects were worsened in neural lineage-specific AhR conditional knockout mice (Nestin-Cre/Ahr-flx, nAhrCKO) mice. In summary, our data suggest that LPS-induced astrogliosis involves the activation of IDO-AhR pathway to negatively feedback the LPS-induced proinflammatory responses, and this mechanism of action might account for the maintenance of cognitive function after peripheral inflammation insult.

Disclosures: Y. Gan: None. **S. Wu:** None. **C. Lin:** None. **Y. Huang:** None. **F. Shie:** None. **H. Lin:** None. **C. Jeng:** None. **Y. Lee:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.13/T3

Topic: B.12. Glial Mechanisms

Support: NIH

NSF GRFP

Title: Layer specific cortical astroglia regulate dendritic branching and spine density

Authors: *S. J. MILLER, J. D. ROTHSTEIN
Neurology/Cellular & Mol. Med., Johns Hopkins Sch. of Med., Baltimore, MD

Abstract: Astrocytes are the most abundant cell type in the CNS, playing essential roles in maintaining the homeostasis, interactions with local microglia and oligodendroglia, synaptic pruning and synaptic generation, and blood brain barrier maintenance. Historically, astrocytes have been classified into two groups: protoplasmic astrocytes of the grey matter and fibrous astrocytes of the white matter. More recently however, groups are unraveling the heterogeneity of astrocytes in different neuroanatomical regions, although the unique functions of these astrocyte subgroups is largely unknown. To better understand astrocyte heterogeneity, we created a transgenic mouse model that retains tdTomato-expression in a well defined neuroatomically limited subset of grey matter astrocytes. In the cortex, these astrocytes are limited to layer 5 and a smaller subset to layer2/3. To understand if these these cells have unique molecular, biochemical and/or physiological properties, we FACs sorted adult Td-Tom positive astrocytes and compared them to the remaining cortical astrocytes. These cells were subjected to microarray and proteomic analyses, following by immunostaining in vivo to validate genes highly expressed and/or unique to these this astrocyte subset. We also were able to study these unique astrocytes in vitro as well as in populations of human iPS derived astrocytes. After extensive 'omic and bioinformatic analyses we uncovered several unique genes to this subset. Interestingly, a selected neuronal gene encodes a secreted protein and the cognate G-protein coupled receptor (GPCR) for this protein was localized to the regional cortical layer 5 specific astrocytes. Next, we began to elucidate this GPCR's signaling pathway in cortical layer V. We found that this astrocyte subset signals through this GPCR by responding to neuronal input and evokes the release of specific polypeptides that act to increase neighboring neuron dendritic branching and spine density. After removal of this astrocyte GPCR and/or its downstream astrocyte specific polypeptide, we see a dramatic effect on spine density in vivo which strongly correlates to phenotypes seen in neural disorders including mental retardation. This is one of the first studies of its kind to unravel a specific astrocyte subset in cortical layer specific signaling, which appear to aid in the regulation and/or development of layer specific neuronal processes and which may act to promote neuronal health.

Disclosures: S.J. Miller: None. **J.D. Rothstein:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.14/T4

Topic: B.12. Glial Mechanisms

Support: National Science Foundation Grant 0924372

Title: Measurement of H^+ fluxes from cultured mouse hippocampal astrocytes using self-referencing H^+ selective microelectrodes

Authors: *J.-I. CHOI[1], R. P. MALCHOW[2]
[1]Biol. Sci., Univ. of Illinois at Chicago, Chicago, IL; [2]Dept. Biol. Sci. & Ophthalmology, Univ. of Illinois at Chicago Dept. of Biol. Sci., Chicago, IL

Abstract: pH regulation in the brain is important and even small changes in intracellular or extracellular pH influence the functions of numerous enzymes and ion channels. Activities such as extracellular glutamate removal process done by glial cells are associated with change in extracellular levels of H+, which can by itself have a potent modulatory effect on neuronal excitability and synaptic transmitter release. In the present study, we have used self-referencing H+-selective microelectrodes to examine standing levels of extracellular H+ from quiescent hippocampal astrocytes cultured from mice and have also examined changes in the level of extracellular H+ that occur upon addition of the different neurotransmitters. Cultured astrocytes recorded in a 24 mM bicarbonate-based saline solution or1 mM HEPES buffered solution exhibit a standing acidic flux. The standing flux observed in the 1 mM HEPES condition remained when all of the extracellular sodium was replaced with choline . Application of glutamate induced a transient extracellular alkalinization, consistent with its transport into the glial cells. Application of adenosine triphosphate (ATP) induced a pronounced extracellular acidification. These results are the first to show extracellular H+ levels adjacent to isolated glial cells using self-referencing electrodes and suggest the possibility that changes in extracellular H+ by glial cells in response to the release of different neurotransmitters may play a role in modulation of activity within the nervous system.

Disclosures: J. Choi: None. **R.P. Malchow:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.15/T5

Topic: B.12. Glial Mechanisms

Support: CONACYT CB-254728

Title: Cathepsin D activity in the mouse hippocampus in various developmental stages, revealed by the cleavage of prolactin into vasoinhibins

Authors: *E. ARNOLD[1], F. MACÍAS[2], R. M. AROÑA[2], C. CLAPP[2], G. MARTÍNEZ DE LA ESCALERA[2]
[1]Catedrática CONACYT-Instituto de Neurobiología, Univ. Nacional Autónoma de México (UNAM), Queretaro, Mexico; [2]Inst. de Neurobiología, Univ. Nacional Autónoma de México (UNAM), Querétaro, Mexico

Abstract: Vasoinhibins are a family of peptides derived from the pituitary hormone prolactin (PRL), named for their inhibiting effects on angiogenesis, vasopermeability and vasodilation. Vasoinhibins are generated via proteolytic cleavage of PRL by several proteases, including cathepsin D, matrix metalloproteases, and bone morphogenic protein-1. Vasoinhibins share the NH2-terminal region of PRL and range in molecular mass between 11 and 18 kDa. PRL and vasoinhibins have been detected in different regions of the central nervous system, where they trigger opposite effects. However little is known about the presence of vasoinhibins in the hippocampus, thus in the present study we explored the activity of the converting enzymes, particularly cathepsin D in the hippocampus from mice in different stages of the life cycle. For that purpose the hippocampus from 16 days old embryos (E16), neonates (N) and adult (A) mice were obtain and lysated in a lysis buffer (1M Tris-HCl, EGTA 0.2M, EDTA 0.2M, 1% Igepal, 0.1M Na_3VO_4, 0.05M NaF, 5mM $Na_4P_2O_7$, 0.26M sucrose). It is well known that astrocytes express and secrete cathepsin D during development and pathological conditions, thus we also isolated astrocytes from hippocampus of E16 and N mice. Astrocytes cultures were lysated in the same buffer. Then, increasing amounts of the hippocampal and astrocytes lysates were incubated with 50 ng of a rat PRL standard in a pH 5 buffer (0.1M Citric acid, 0.1M Na_2HPO_4, NaCl 0.15 M) during 24h at 37°C in the presence or absence of the enzymatic inhibitor pepstatin A, that inhibits the action of cathepsin D. The levels of PRL and vasoinhibins were measured by immunoblotting. Our results show vasoinhibins in all the samples, suggesting the hippocampal expression of enzymes that cleaves PRL into vasoinhibins at the three stages of development tested. A hippocampal cathepsin D cleaves PRL into a vasoinhibin of 16kDa in the three stages analyzed. Moreover, we found that neonatal astrocytes posses higher cathepsin D proteolytical activity in comparison with embryonic astrocytes lysates, since lower concentrations of lysates are able to cleavege PRL into vasoinhibins. Altogether these findings show that a regulation of the PRL-vasoinhibins axis operates in the hippocampus of the mice, and that it is developmental-related to the enzymatic activity of cathepsin D in astrocytes.

Disclosures: E. Arnold: None. **F. Macías:** None. **R.M. Aroña:** None. **C. Clapp:** None. **G. Martínez de la Escalera:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.16/T6

Topic: B.12. Glial Mechanisms

Support: NIH-NIGMS-SC1-GM088019

NIH-NINDS-R01-NS065201

NIH-NIMHD-G12-MD007583

NIH-NIGMS-R25-GM110513

Title: KIR4.1 potassium channel in hyperglycemic conditions: Downregulation via MIR-205 overexpression in astrocytes

Authors: *D. E. RIVERA-APONTE[1], M. P. MENDEZ-GONZALEZ[2,4], K. MELNIK-MARTINEZ[3], C. MALPICA-NIEVES[2], F. TEJEDA-BAYRON[2], S. SKATCHKOV[3], M. J. EATON[1]
[1]Biochem., Univ. Central Del Caribe, Bayamon, PR; [2]Biochesmitry, [3]Physiol., Univ. Central del Caribe, Bayamon, PR; [4]Natural Sci., Univ. of Puerto Rico Aguadilla, Aguadilla, PR

Abstract: Astrocytes play a critical role in protecting neurons by maintaining extracellular homeostasis, preventing neurotoxicity and regulating the neuronal network through glutamate uptake, potassium buffering and gliotransmitter release. These functions are supported by the presence of potassium channels, mainly by Kir4.1 inwardly rectifying potassium channels, in the membranes of astrocytes and other glial cells. Hyperglycemia affects patients who suffer from diabetes and its effects on the central nervous system could be critical since astrocytes grown in hyperglycemic conditions have decreased levels of Kir4.1 potassium channels as well as impaired potassium and glutamate uptake. The link between hyperglycemic condition and functional downregulation of Kir4.1 is still missing, however previous studies performed in human corneal epithelial cell injury demonstrated that regulation of Kir4.1 expression occurs via microRNAs, specifically miR-205. The aim of the present study is to test 1) if astrocytes express miR-205 and 2) if miR-205 regulates Kir4.1 expression under hyperglycemic conditions. We used q-PCR to assess the levels of miR-205 in astrocytes grown in high glucose (25 mM) DMEM compared to control astrocytes grown in normal glucose (5 mM). We found that not only was miR-205 expressed in astrocytes, but its expression was increased when astrocytes were grown in hyperglycemic conditions. Astrocytes grown in high glucose had a 6-fold up-regulation of miR-205 levels compared to astrocytes grown in normal glucose condition. Transfection of miR-205 mimic and inhibitor was performed to alter the levels of miR-205 in astrocytes followed by Western blot to assess Kir4.1 channel levels in these cells. Astrocytes treated with mimic miR-105 had a 32.7% reduction of Kir4.1 protein levels compared to control (mock-transfected) cells. In contrast, astrocytes transfected with inhibitor miR-205 were significantly up-regulated compared to mock 39%. Taken together, our data indicate that miR-205 negatively regulates the expression of Kir4.1 in astrocytes during high glucose conditions.

Disclosures: D.E. Rivera-Aponte: None. **M.P. Mendez-Gonzalez:** None. **K. Melnik-Martinez:** None. **C. Malpica-Nieves:** None. **F. Tejeda-Bayron:** None. **S. Skatchkov:** None. **M.J. Eaton:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.17/T7

Topic: B.12. Glial Mechanisms

Support: Rett Syndrome Foundation for Basic Research Grant (#2916)

National Institutes of Health (grant number R01 NS075062)

UAB CCTS (grant number 5UL1RR025777)

Civitan International Research Center

Title: Kir4.1, a novel target for MeCP2

Authors: *U. KAHANOVITCH[1], V. A. CUDDAPAH[2], N. L. PACHECO[2], L. M. HOLT[1,2], A. K. PERCY[3], M. L. OLSEN[4]
[1]Sch. of Neurosci., Virginia Tech., Blacksburg, VA; [2]Cell, Developmental and Integrative Biol., [3]Pediatrics, Civitan Intl. Res. Ctr., Univ. of Alabama at Birmingham, Birmingham, AL; [4]Virginia Tech, Sch. of Neurosci., Blacksburg, VA

Abstract: Rett Syndrome (RTT) is an X-linked neurodevelopmental disorder typified by apparently normal development until 6-18 months of age, when motor and communicative skills regress and hand stereotypies, autonomic dysfunction, and seizures present. Over 95% of all reported RTT is caused by spontaneous mutations in a single gene, methyl-CpG-binding protein 2 (MeCP2). Nearly all RTT research has focused on neuronal dysfunction in RTT, however, it was recently demonstrated that restoration of MeCP2 function selectively to astrocytes reversed several deficits in a murine model of RTT, suggesting astrocytes contribute to the RTT phenotype. The mechanism of this rescue is unknown. Here we demonstrate that Kir4.1, a glia-specific inward-rectifying potassium channel that mediates several critical astrocyte membrane properties is disrupted in MeCP2 deficient mice. Astrocytes from MeCP2-deficient mice express significantly less Kir4.1 mRNA and protein, which corresponds with a 50% reduction in Ba^{2+}-sensitive Kir4.1 mediated currents. Indeed, Kir4.1 protein and mRNA expression is significantly reduced well before symptom onset. ChIP analysis revealed a direct molecular interaction between MeCP2 and the Kir4.1 gene promoter in WT animals, an interaction that is lost in MeCP2 deficient mice. These are the first data implicating a direct molecular target of MeCP2 in astrocytes and provide novel mechanistic insight explaining how astrocytic dysfunction may contribute to RTT. Future work will concentrate on elucidating the role of MeCP2 loss in the brainstem in the breathing dysfunction phenomenon in RTT.

Disclosures: **U. Kahanovitch:** None. **V.A. Cuddapah:** None. **N.L. Pacheco:** None. **L.M. Holt:** None. **A.K. Percy:** None. **M.L. Olsen:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.18/DP04/T8 (Dynamic Poster)

Topic: B.12. Glial Mechanisms

Support: NSF IOS 1354913

NIH U01 MH109062 BRAIN

Title: Glial heterogeneity in the molecular layer of hippocampal dentate gyrus

Authors: *G. NASERI KOUZEHGARANI[1,2], M. U. GILLETTE[1,2,3]
[1]Neurosci. Program, [2]Beckman Inst. for Advanced Sci. & Technol., [3]Dept. of Cell & Developmental Biol., Univ. of Illinois at Urbana-Champaign, Urbana, IL

Abstract: Glia are emerging as heterogeneous elements within the molecular layer of the dentate gyrus (ML-DG). Astrocytes, which display linear voltage-current (IV) curves, are coupled through gap junctions, whereas oligodendrocyte precursor cells (NG2+) exhibit voltage-dependent currents and lack gap-junction coupling. Little is known about the morphological, electrophysiological, or coupling dynamics of these glial cells with regards to the time of day. Using whole-cell patch-clamp recording, sulforhodamine-B, a gap-junction permeable dye, was injected to a single cell in morning or evening, and its spread to other cells was analyzed along with identifying cell-specific markers. Astrocytes displayed linear IV profiles, as expected, with very low resistances (< 20 MΩ) at both times-of-day, but the number of cells coupled as a cluster was significantly higher during the evening. In the case of NG2 cells, sulforhodamine-B was not transferred to other cells, indicating the lack of coupling. Cells with two different types of morphologies with distinct membrane properties were observed: (1) isolated cells displayed a non-linear IV curve with a high resistance (250-350 MΩ), and (2) NG2-NG2 pairs with direct somatic contact and extended processes in opposite directions. These cells, termed 'NG2 doublets', exhibited linear IV profiles with lower resistances (< 100 MΩ). No coupling was found between NG2 cells and astrocytes. Immunohistochemistry with double staining of the recorded slices post-fixation confirmed the identity of the injected cell as either astrocyte (GFAP-positive, non-radial glia) or oligodendrocyte precursor cell (NG2-positive). Quantification of NG2 cells at the two timepoints provides insights into the cell dynamics. Thus, within hippocampal ML-DG, we find different morphological, electrophysiological, and coupling properties of two types of glia with respect to the time of day.

Disclosures: G. Naseri Kouzehgarani: None. **M.U. Gillette:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.19/T9

Topic: B.12. Glial Mechanisms

Title: Establishment of a direct induction method toward astrocytic cells using non-viral polycistronic vector as a neurological disease modeling platform

Authors: *R. TOMOOKA[1], Z. ZHOU[2], T. SANOSAKA[1], S. BANNO[1], I. KOYA[1], M. CHAI[1], R. SHIMAMURA[1], T. ANDO[1], H. OKANO[1], J. KOHYAMA[1]
[1]Keio Univ. Sch. of Med., Tokyo, Japan; [2]E-WAY Res. Laboratory, Discovery, Med. Creation, Neurol. Business Group, Tsukuba, Japan

Abstract: It has becoming apparent that glial cells play pivotal roles in the pathogenesis of neurodegenerative disease. To achieve pathologic elucidation and innovative drug development targeting the disease, it is useful to establish disease-in-a-dish model. However, there is few robust methodology for the rapid and efficient preparation of disease-specific astrocytic cells. Here, we estabslished a polycistronic episomal expression vector which expressed astrocyte-inducing factors using the self-cleavage 2A peptide. To validate this system, we introduced the vector onto human neural progenitors, and efficiently generated astrocytic cells. Furthermore, as a proof-of-concept trial, we generated astrocytic cells from Alzheimer's disease patient-derived cells and performed functional analyses and transcriptome analysis. We found the pathways associated with inflammatory signaling were upregulated in Alzheimer's disease-patient astrocytes, adding new layers of pathogenesis of Alzheimer's disease involving neuroinflammatory response evoked by astrocytes.

Disclosures: R. Tomooka: None. **Z. Zhou:** None. **T. Sanosaka:** None. **S. Banno:** None. **I. Koya:** None. **M. Chai:** None. **R. Shimamura:** None. **T. Ando:** None. **H. Okano:** None. **J. Kohyama:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.20/T10

Topic: B.12. Glial Mechanisms

Support: NIH Grant R01GM102756

NIH Grant R01GM102756

NIMH Grant MH099587

NIMH Grant MH100031

NICHD Grant HD076892

NICHD Grant HD03352

NIH T32 Fellowship GM007215

Title: Functional and molecular characterization of human pluripotent stem cell-derived regional astrocytes

Authors: *R. A. BRADLEY[1], J. SHIREMAN[2], C. MCFALLS[2], J. CHOI[2], Y. DONG[3], S. G. CANFIELD[4], M. CHIANG[6], J. JONES[7], A. PETERSEN[5], S. PALACEK[2], E. SHUSTA[2], C. KENDZIORSKI[2], Y. YANG[8], S.-C. ZHANG[1]
[1]Neurosci., [2]Univ. of Wisconsin - Madison, Madison, WI; [3]Waisman Ctr., Univ. of Wisconsin, Madison, WI; [4]Chem. and Biol. Engin., [5]Neurosci., Univ. of Wisconsin Madison, Madison, WI; [6]Tufts Univ., Boston, MA; [7]Univ. of Wisconsin-Madison Wasiman Ctr., Madison, WI; [8]Neurosci., Tufts Univ. Sch. of Med., Boston, MA

Abstract: Astrocytes are known to exhibit different morphologies and functions in different parts of the brain and spinal cord. The origin of this molecular and functional heterogeneity is largely unknown. Utilizing astrocytes generated from regionally-specified neural progenitors derived from pluripotent stem cells, we analyzed the transcription profiles by RNA sequencing and assessed the functional properties and effects of regional astrocytes on neurons and blood-brain barrier formation. We found distinct molecular profiles that are associated with each region, including predicted homeodomain transcription factors as well as transcripts which suggest different functional properties. Functional analysis of these regional astrocytes revealed differences in membrane potential and calcium signaling as well as differential effects on neurite outgrowth, synaptic formation and blood-brain barrier permeability in co-culture models. These results suggest that differences in regional astrocytes and their functional properties are partly attributed to their developmental origins.

Disclosures: R.A. Bradley: None. **J. Shireman:** None. **C. McFalls:** None. **J. Choi:** None. **Y. Dong:** None. **S.G. Canfield:** None. **M. Chiang:** None. **J. Jones:** None. **A. Petersen:** None. **S. Palacek:** None. **E. Shusta:** None. **C. Kendziorski:** None. **Y. Yang:** None. **S. Zhang:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.21/T11

Topic: B.12. Glial Mechanisms

Support: Strategic Research Foundation Grant-aided Project for Private Universities from Ministry of Education, Culture, Sport, Science, and Technology, Japan (MEXT), 2014-2018 (S1411003)

Title: Spatiotemporal modulation of neuronal activity by astrocytic second phase calcium oscillation

Authors: *T. KIKUCHI, Y. SHINDO, K. HOTTA, K. OKA
Keio Univ., Yokohama, Kanagawa, Japan

Abstract: The relative number of astrocytes to neuronal number increases dramatically in association with the brain complexity. It is plausible that the large number of astrocytes with brain evolution could be a function of network complexity; in fact, increasingly dense and sophisticated synaptic networks require greater degrees of local modulation and fine control by astrocytes. Recent studies show that astrocytes are actively participating to synaptic transmission by releasing transmitters. Their spatiotemporal dynamic involvement in neural plasticity is now widely accepted; however, details of gliotransmission associated with calcium increase are still unclear. We report the first evidence that astrocytes show second phase calcium oscillation, which is minute order and long-lasting, by glutamate stimulation. Interestingly, this second phase oscillation makes certain “firing pattern” in the global field. We also uncover P2 purinergic receptor antagonists, suramin, inhibits this oscillation, suggesting that ATP release from astrocytes provokes second phase calcium oscillation. The lower cell density also suppresses significantly their second phase calcium oscillation, indicating that cycle of this oscillation depends on the quantity of receivable ATP released from neighboring astrocytes. To further pursue the mechanism of this event, we constructed a FRET-based ATP indicator, which can be localized specifically outside of cell membrane, named outATeam3.10. We combined two existing genetically encoded fluorescent sensors: ATeam3.10 for the ATP sensing compound and iGluSnFR for the extracellular membrane localization tag. This sensor has linearity in the range between 0.1 μM to 20 μM ATP. Using this newly created outATeam3.10 and R-GeCO, we successfully recorded simultaneous imaging of astrocytic ATP release and the temporal change of second phase calcium oscillations. To further understand the spatiotemporal astrocytes’ implication for CNS, our method for real-time monitoring of ATP levels outside of individual living cells could be one useful approach. The long-lasting second phase calcium oscillation in

astrocytes might be explained by spatiotemporal gliotransmission modality; and hence, they probably involve in the regulation of neural activity.

Disclosures: **T. Kikuchi:** None. **Y. Shindo:** None. **K. Hotta:** None. **K. Oka:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.22/T12

Topic: B.12. Glial Mechanisms

Support: IGERT 0965918

STC EBICS CBET 0939511

DBI 1450962

MH101655

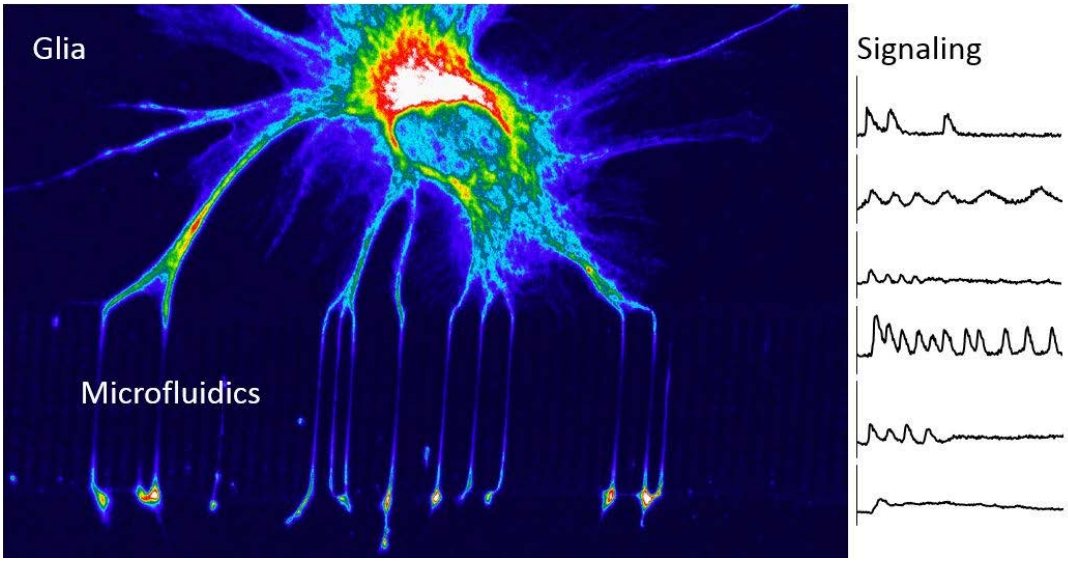
University of Tennessee-Knoxville

Title: Focal stimulation and network activation of astrocytes and glial networks using microfluidics

Authors: ***L. J. MILLET**[1], A. JAIN[2], M. U. GILLETTE[3]
[1]Joint Inst. Biol. Sci., Univ. of Tennessee., Oak Ridge, TN; [2]Dept. of Cell and Develop. Biol., Univ. of Illinois at Urbana-Champaign, Urbana, IL; [3]Dept. of Cell & Developmental Biol., Univ. of Illinois, Urbana, IL

Abstract: Glia function to maintain homeostasis, myelinate neurons, recycle neurotransmitters, modulate neuronal function, and shuttle energy to neurons. Astrocytic glia coordinate with neighboring glia, neurons and endothelial cells through cellular extensions that form functional cellular domains. Here we investigate how individual astrocytic processes respond to chemical cues and propagate calcium signals within the cell and throughout glial networks. We create microenvironments that enable controlled fluidic domains that are amenable for stimulating glial appendages without biasing the network to the stimuli. Nanoliter-scale microfluidic cell culture and imaging platforms are fabricated through a novel, sequential process that involves solvent-extracted polydimethylsiloxane (PDMS) and hydrothermal annealing. Primary glial cells from the cerebral cortex are cultured within this platform to enable focal application and imaging of sub-cellular calcium transients. Using immunochemistry and confocal microscopy, we evaluate glial morphology and find that glia extend fine, micron-scale processes into neighboring domains. Sub-cellular and network level calcium transients are imaged using Fluo-4 AM and a

Zeiss LSM-510 Meta NLO system equipped with an argon laser. The activation of calcium transients is achieved by focal application of ATP and glutamate, Ca^{2+} wave propagation occurs at a rate of 11-12 micron/sec. Repeated elevated Ca^{2+} transients in distant glial cells are also achieved; individual responses and sustained oscillations show prototypical morphology and are reminiscent of microburst activity. Our findings extend previous studies of glial signaling to evaluate local signaling at single processes that sense, respond to, and transmit information regarding environmental cues. Our approach demonstrates a simple, yet powerful system to interrogate the activity of individual cells and networks of glia in an unprecedented way using microfluidics.



Disclosures: **L.J. Millet:** None. **A. Jain:** None. **M.U. Gillette:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.23/U1

Topic: B.12. Glial Mechanisms

Support: NIH/NIDA DA032681

NIH/NIDA DA031747

NIH/NIDA DA041513

Title: Dopamine exposure induces phenotypic changes in rat hippocampal astrocytes

Authors: *A. GALLOWAY[1], A. ADELUYI[2], S. MATHEW[2], J. R. TURNER[2], P. I. ORTINSKI[1]
[1]Pharmacology, Physiology, and Neurosci., Univ. of South Carolina Sch. of Med., Columbia, SC; [2]Drug Discovery and Biomed. Sci., Univ. of South Carolina, South Carolina Col. of Pharm., Columbia, SC

Abstract: Dopamine, a major neuromodulator, is critical for processing of reward and is implicated in etiology of drug addiction. The effects of dopamine on glial cells have been historically understudied, yet research indicates that astrocytes express dopamine receptors and respond to dopaminergic neuron activity. Using an *in vitro* model of elevated extracellular dopamine, our evidence reveals a rapid dopamine-induced switch from fibroblast-like to stellated morphology in cultured rat hippocampal astrocytes. A one hour exposure to dopamine (75µM) produced a pronounced increase in the number of primary astrocyte processes which was accompanied by a decrease in GFAP-positive area. Exposure to antioxidants N-acetylcysteine or sodium metabisulfite in the presence of extracellular dopamine did not disrupt dopamine effects on astrocyte morphology. However, treatment with dopamine receptor antagonists significantly blunted dopamine-induced stellation. Morphological changes were associated with a decrease in acetylated tubulin, consistent with decreased GFAP-positive area. RNA-sequencing revealed that a number of immediate early genes (IEGs) previously linked to structural and functional adaptations in neurons are also differentially expressed in dopamine-treated astrocytes. At the functional level, dopamine treatment had no effect on the amplitude of astrocytic intracellular Ca^{2+} transients, but increased Ca^{2+} wave duration. Together these findings indicate that elevated extracellular dopamine induces phenotypic changes in cultured astrocytes. Physiological implications for astrocyte-mediated dopamine effects in the context of drug addiction remain to be examined.

Disclosures: A. Galloway: None. **A. Adeluyi:** None. **S. Mathew:** None. **J.R. Turner:** None. **P.I. Ortinski:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.24/U2

Topic: B.12. Glial Mechanisms

Support: UL Lafayette Undergraduate Research Mini Grant

NARSAD YI award

UL Lafayette Dean Start Up Funds

Title: Fibroblast growth factor receptor 1 expression in animal models of chronic stress and injury

Authors: *K. M. SMITH[1], J. C. COLLETTE[2], H. TORRES[2], L. B. RUBIN[2]
[2]Biol., [1]Univ. of Louisiana At Lafayette, Lafayette, LA

Abstract: Fibroblast Growth Factor Receptor 1 (FGFR1) is an important regulator of cortical development, function, and response to injury. We have studied the cellular expression of the Fgfr1 gene by employing the *tgFgfr1-EGFP* mouse line. This BAC transgenic line was generated by the GENSAT project, and our previous studies demonstrated that GFP is expressed in a manner that is consistent with known roles of FGFR1 in development and function. In development, GFP expression was found in telencephalic radial glia, hippocampal anlage, and rhombic lip where it is known to play roles in stem cell maintenance and amplification; it was also expressed in midline radial glia, where FGFR1 is crucial to the formation and migration of midline radial glial astrocytes; and in Bergmann Glia, where FGFR1 and FGFR2 signaling cooperate to maintain Bergmann Glia morphology and attachment to the pial membrane. In adult mice, we also found expression in GFAP positive astrocytes throughout the brain, Olig2+ cells, a minority of cortical and hippocampal neurons, BLBP+ cells of the lateral subventricular zone, and hippocampal DG and CA regions, as well as the hypothalamus. Here, we investigate whether the *tgFgfr1-EGFP* mouse line can be used to investigate changes in GFP expression after chronic unpredictable stress (CUS), and in response to injury. Our purpose was to validate whether this model responds to manipulations that have previously been reported to lead to changes in *Fgfr1* expression. We found that chronic unpredictable stress leads to upregulation of *Fgfr1* in the CA region of the hippocampus. The injury model we chose to study was the cuprizone model of demyelination, which is achieved by ingestion of 0.2% cuprizone in the feed. We found that *Fgfr1* driven GFP was upregulated in the recovery period of chronic cuprizone administration. Interestingly, this upregulation of GFP in the corpus callosum was colocalized with GFAP, indicating that the upregulation in the corpus callosum was due to increased numbers of astrocytes expressing *Fgfr1*, as opposed to oligodendrocyte or oligodendrocyte precursors. Current and future studies will investigate whether inactivation of *Fgfr1* in postnatal astrocytes leads to impaired injury response to demyelination injury, or changes in astrocyte and stem cell function.

Disclosures: **K.M. Smith:** None. **J.C. Collette:** None. **H. Torres:** None. **L.B. Rubin:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.25/U3

Topic: B.12. Glial Mechanisms

Support: NSF GRFP

Title: Calcium signals in LC axons and astrocytes in the hippocampus during navigation

Authors: *A. KAUFMAN, W. LI, J. BOWLER, E. M. BALOUGH, A. LOSONCZY
Columbia Univ., New York, NY

Abstract: Astrocytes exhibit increases in calcium in response to certain neurotransmitters. Astrocytes have been shown to respond to norepinephrine in barrel cortex, cerebellum, and visual cortex. Using a two-photon microscope, we are simultaneously imaging catecholaminergic axons projecting from the locus coeruleus and astrocytes in the hippocampus of awake, behaving mice. We are examining the correlation between astrocyte calcium signals and axon calcium signals during goal-oriented reward learning on a cued treadmill belt, as well as when these signals occur in relation to behavior. We are examining both specific behavioral epochs such as the start of running and reward-seeking behavior, and larger behavioral events such as learning the location of the reward. We can compare this to random foraging for reward on a cued belt, in which the animals do not need to learn the location of a reward. In addition, we are characterizing the responses of both the axons and astrocytes during the presentation of stimuli of positive, negative and neutral valences.

Disclosures: A. Kaufman: None. **W. Li:** None. **J. Bowler:** None. **E.M. Balough:** None. **A. Losonczy:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.26/U4

Topic: B.12. Glial Mechanisms

Support: NSFC grant 31330024

NSFC grant 31171026

Title: Stretch induced ATP release in hippocampal astrocytes

Authors: *Z. ZHOU[1], Y. XIONG[2,3,4,4], S. TENG[2], S. SUN[2], N. GUO[2], N. GUO[2], M. LI[2], F. ZHU[2], C. WANG[2], L. ZHENG[2], Z. RAO[3]
[1]Inst. Mol. Med, Peking Univ., Beijing, China; [2]Inst. of Mol. Med., Beijing, China; [3]Inst. of Neurosciences, Xi'an, China; [4]Affiliated Hosp. of Air Force Inst. of Aeromedine, Beijing, China

Abstract: Astrocytic ATP release is essential for brain functions such as synaptic long term potentiation for learning and memory. However, whether/how ATP is released via exocytosis

remains in hot debates. Most of previous studies on non-vesicular ATP releases are studied with relative indirect procedures. In contrast, two recent works report vesicular ATP release using more direct assays. In the present work, we re-investigated astrocytic ATP release at single vesicle resolution in cultured hippocampal astrocytes. Following a mechanical stretch stimulation (Mstim) on a single astrocyte we studied Ca^{2+}*-dependence of* ATP releases, which are in comparison to Ca^{2+}*-dependent* quantal ATP release in chromaffin cells. The astrocytic ATP release did not require lysosome exocytosis but was inhibited by either selective-antagonist or genetic knock-down of a nonselective cation channel. In summary, there is a Mstim-induced ATP release through a cation channel in hippocampal astrocytes.

Disclosures: Z. Zhou: None. **Y. Xiong:** None. **S. Teng:** None. **S. Sun:** None. **N. Guo:** None. **N. Guo:** None. **M. Li:** None. **F. Zhu:** None. **C. Wang:** None. **L. Zheng:** None. **Z. Rao:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.27/U5

Topic: B.12. Glial Mechanisms

Support: Foerster-Bernstein Postdoctoral Fellowship

The Hartwell Foundation Postdoctoral Fellowship

Simons Foundation Autism Research Initiative Pilot Award

Title: Identification of a novel regulator of astrocyte development and maturation

Authors: *K. T. BALDWIN[1], J. A. STOGSDILL[1], R. ESTEVEZ[2], C. EROGLU[1]
[1]Dept. of Cell Biol., Duke Univ., Durham, NC; [2]Departament de Ciències Fisiològiques II, Univ. of Barcelona, Barcelona, Spain

Abstract: Astrocytes, the major glial cells of the brain, play critical roles in a wide variety of brain functions, including synapse formation, ion homeostasis, and formation of the blood-brain barrier. The functional complexity of astrocytes is reflected in their elaborately branched morphology. Astrocytes form extensive arbors terminating in thousands of fine processes that structurally and functionally interact with synapses. Despite the vital role of astrocytes in the brain, the cellular and molecular mechanisms that drive astrocyte development are poorly understood. Furthermore, it is unclear how disruptions to astrocyte development affect astrocyte function and drive disease pathogenesis. To address these critical knowledge gaps, we performed a candidate screen of astrocyte-enriched, disease-linked genes to identify novel regulators of astrocyte development and maturation. This screen identified the cell adhesion molecule

HepaCAM (also known as GlialCAM) as a critical regulator of astrocyte morphological development, both in vitro and in vivo. Knockdown of HepaCAM severely reduces astrocyte morphological complexity in astrocyte-neuron co-cultures, and impairs astrocyte development in mouse visual cortex. In humans, mutations to the extracellular domain of HepaCAM cause developmental brain disorders, namely Megalencephalic Leukoencephalopathy with Subcortical Cysts (MLC), and macrocephaly with intellectual disability and autism. Ongoing studies are investigating how these disease mutations alter astrocyte development and contribute to astrocyte dysfunction that is associated with disease pathogenesis.

Disclosures: K.T. Baldwin: None. **J.A. Stogsdill:** None. **R. Estevez:** None. **C. Eroglu:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.28/U6

Topic: B.12. Glial Mechanisms

Title: Ng2 knock-out in the neu7 reactive astrocyte cell line to promote neuronal regeneration

Authors: *S. M. CIOMBOR, A. HAY, M. HABERMAN, M. FALLERT, D. R. COOK-SNYDER
Carthage Col., Kenosha, WI

Abstract: Stroke remains a leading cause of death and disability worldwide. After an ischemic stroke, neuronal regeneration is prohibited due to the astrocytes surrounding the tissue becoming reactive. Reactive astrocytes are non-permissive to axon outgrowth, and improving our understanding of astrocyte reactivity can facilitate development of more targeted therapies for neuronal regeneration. Neu7s are an astrocyte cell line that models this reactive phenotype due to an increased production of inhibitory chondroitin sulfate proteoglycans. Of these proteoglycans, chondroitin sulfate proteoglycan 4 (NG2) has been found to be the most inhibitory to axon regeneration. A7 cells are a model cell line for astrocytes that are permissive to axon outgrowth, having a downregulated production of NG2. The CRISPR/Cas9 system was used to produce NG2 knockout Neu7 and A7 cell lines. NG2 is a 35,046 base pair long gene with ten exons. We created guide sequences targeting exons 1, 3, 5 and 10. Bacterial cloning was used to construct plasmids containing guide sequences, Cas9 and green fluorescent protein (GFP). Sanger sequencing and alignment identified correctly constructed plasmids. Neu7 cells were transfected with plasmids containing NG2 guide sequences. Ongoing experiments will continue to determine conditions to increase transfection efficiency, transfect A7 cells, identify unique cellular markers, and develop neuronal co-cultures with transfected astrocytes. These co-cultures will help assess the role of NG2 in axon outgrowth inhibition. We believe these experiments will aid in the development of new therapies for neuronal regeneration.

Disclosures: **S.M. Ciombor:** None. **A. Hay:** None. **M. Haberman:** None. **M. Fallert:** None. **D.R. Cook-Snyder:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.29/U7

Topic: B.12. Glial Mechanisms

Support: NIH F32EY028028

Title: The role of astrocytic GABA transport during sensory processing in visual cortex

Authors: *G. O. SIPE[1], R. GARCIA[2], R. V. RIKHYE[2], J. C. PETRAVICZ[2], M. SUR[2]
[2]Dept. of Brain and Cognitive Sci., [1]MIT, Cambridge, MA

Abstract: Increasing evidence indicates that astrocytes actively modulate neuronal network activity through dynamic neurotransmitter clearance at the synaptic cleft. Although neurotransmitter clearance maintains synaptic homeostasis, it can also affect synaptic efficacy and subsequent information processing. Most work on astrocytic neurotransmitter clearance focuses on glutamate uptake via the transporters GLT-1 and GLAST, however astrocytes also express the GABA transporters GAT-1 & GAT-3. In particular, multiple studies have indicated that GAT-3 is selectively expressed in astrocyte processes near synaptic clefts and is thought to mediate tonic inhibitory states. Though GAT-3 blockade has been shown to delay the onset of seizure events in epilepsy models, it is not known how GAT-3 activity influences cortical information processing. To explore this question, we investigated the function of GAT-3 in primary visual cortex of the adult mouse. We characterized GAT-3 function in the cortex using immunohistochemistry and confirmed previous reports that GAT-3 expression heavily co-localizes with astrocytic processes and displays layer-specific heterogeneity. We then characterized GAT-3 function in the visual cortex using a combination of 2-photon microscopy, optogenetic activation, and slice electrophysiology. Our data suggests that neuronal reliability and tuning properties are disrupted with GAT-3 blockade. In conjunction with previous work in the lab investigating the role of astrocytic GLT-1 in visual cortex, these data indicate that astrocytes contribute to the crucial balance of excitation and inhibition necessary for cortical information processing.

Disclosures: **G.O. Sipe:** None. **R. Garcia:** None. **R.V. Rikhye:** None. **J.C. Petravicz:** None. **M. Sur:** None.

Poster

384. Astrocytes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 384.30/U8

Topic: B.12. Glial Mechanisms

Support: NIH Grant R01NS062784

NIH Grant R56NS097672

Title: Ultrastructural association of astrocytes with other constituents in the brain

Authors: *C. M. KIYOSHI[1], A. T. TAYLOR[1], K. N. BONI[2], S. A. GARCIA SERRANO[2], R. KUMAR[3], A. N. DRAKE[3], A. M. HEGAZI[1], Y. DU[1], B. MA[1], W. WANG[4], D. TERMAN[2], M. ZHOU[1]
[1]Dept. of Neurosci., The Ohio State Univ. Wexner Med. Ctr., Columbus, OH; [2]Dept. of Mathematics, The Ohio State Univ., Columbus, OH; [3]Metro Early Col. High Sch., Columbus, OH; [4]Dept. of Physiol., Huazhong Univ. of Sci. and Technol., Wuhan, China

Abstract: Astrocytes are extensively connected with each other through gap junctions to form the largest syncytium. The astrocyte syncytium is increasingly recognized to function as a system in the critical homeostatic and signaling roles to distribute energy substrates and regulate interstitial ion and neurotransmitter levels for optimal synaptic transmission. However, the involved ultrastructural basis that underlies these syncytial functions remains largely unclear. In addition, it has not been resolved if the general cytoarchitecture of astrocytes extend in a tree-like (arborization) manner, as is the case for other CNS cells, or in a unique spongiform branching pattern that self-connect through reflexive gap junctions. We used an innovative technique in correlative confocal microscopy and serial block-face scanning electron micrography (SBFEM) to resolve, for the first time, the entire ultrastructure of coupled astrocytes in the hippocampus. We show that astrocytes exhibit a predominantly spongiform morphology that enwrap neural processes. We also show two neighboring astrocytes make multiple parallel contacts only at their fine processes. Mathematical modeling suggests this contact patterning, in part, explains the low resistance pathway between astrocytes. These multiple parallel processes also exist in specialized astrocyte end-feet processes that establish contact with blood vessels. Our data favor the view that astrocytes adopt a spongiform cytoarchitecture with extensive reflexive processes, and establish contacts with other cells with parallel terminal processes. These results are expected to expand our understanding of the structure-function relationship of astrocytes, the astrocyte syncytium, and its role in brain function and disease.

Disclosures: **C.M. Kiyoshi:** None. **A.T. Taylor:** None. **K.N. Boni:** None. **S.A. Garcia Serrano:** None. **R. Kumar:** None. **A.N. Drake:** None. **A.M. Hegazi:** None. **Y. Du:** None. **B. Ma:** None. **W. Wang:** None. **D. Terman:** None. **M. Zhou:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.01/U9

Topic: C.01. Brain Wellness and Aging

Title: Estimation of metabolic energy balance in neurons by using fluorescent imagings

Authors: *M. IIZUMI, R. SUZUKI, R. YAMANAKA, Y. SHINDO, K. HOTTA, K. OKA
Keio Univ., Yokohama-Shi, Japan

Abstract: Animal cells including neurons produce ATP by two pathways: anaerobic glycolysis and oxidative phosphorylation. Pyruvate is located at a branch point of two pathways and lactate is the end product of anaerobic glycolysis. Thus, simultaneous measurement of lactate and pyruvate concentrations allows the estimation of metabolic energy balance between these two pathways. We expect that estimation of the balance between two pathways sequentially and simultaneously would reveal the optimal energy strategy in individual neurons, which are required for heavy information tasks with high metabolic energy consumption. To measure the concentrations of lactate and pyruvate in a single cell with real-time and high temporal resolution, we used genetically-encoded Forester Resonance Energy Transfer(FRET)-based lactate and pyruvate sensors (San Martin *et al*.,2013; San Martin *et al*.,2014). Both of these two sensors use mTFP and Venus as FRET pair fluorescent proteins, we therefore changed recombining FRET pair of pyruvate sensor to mOrange and mCherry for simultaneous imaging in single cells. After recombination, we evaluated FRET pair-changed pyruvate sensor in vitro. FRET pair-changed pyruvate sensor had two peaks for mOrange and mCherry at 565 nm and 610 nm, respectively. Application of pyruvate caused an increase in mOrange fluorescent intensity and decrease in mCherry fluorescent intensity. And exposure to higher concentrations of pyruvate showed that FRET pair-changed pyruvate sensor increased its FRET efficiency, and we confirmed that FRET pair-changed pyruvate sensor worked normally in vitro. In conclusion, we recombined FRET pair of pyruvate sensor and this allows us dual imaging of lactate and pyruvate in the same cell simultaneously. We expect dual imaging of pyruvate and lactate would also reveal the process of cell death in neurodegenerative disease cells.

Disclosures: **M. Iizumi:** None. **R. Suzuki:** None. **R. Yamanaka:** None. **Y. Shindo:** None. **K. Hotta:** None. **K. Oka:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.02/U10

Topic: C.01. Brain Wellness and Aging

Support: NHMRC 1044007

Title: Impairments in both brain and liver glucose metabolism are restored by fructose 1,6-bisphosphate administration in the chronic pilocarpine-SE mouse model

Authors: *T. MCDONALD[1], M. P. HODSON[2], K. BORGES[1]
[1]Univ. of Queensland, St Lucia, Australia; [2]Australian Inst. for Bioengineering and Nanotechnology, Univ. of Queensland, St Lucia, Australia

Abstract: There is growing evidence that suggests that metabolic disturbances contribute the development and progression of seizures. Here we aimed to elucidate which pathways involved in brain glucose metabolism are impaired during the chronic stage of the pilocarpine-status epilepticus (SE) model of epilepsy, and how the treatment of fructose 1,6-bisphosphate (F16BP) altered this metabolism.
Mice were injected with F16BP (1 g/kg, i.p) daily for 6 days, beginning two weeks after pilocarpine-induced SE. Mice were then injected with [U-^{13}C] glucose (558mg/kg, i.p) and sacrificed 15 minutes later. The percent enrichment of carbon-13 was measured in metabolites of the hippocampal formation and liver using LCMS/MS and GCMS, and enzyme activities were measured via spectroscopy.
Mice that developed SE had a reduction in the percent ^{13}C enrichment in most glycolytic intermediates (17-22%, $p<0.05$) in the hippocampal formation, with no change in glycolytic enzyme activities. This suggests that glucose uptake is impaired. Furthermore, the incorporation of ^{13}C in the TCA cycle intermediates was reduced (17-35%, $p<0.05$), coupled with 33% and 55% loss in the activities of pyruvate dehydrogenase and 2-oxoglutarate dehydrogenase, respectively ($p<0.05$ for both). Together this indicates that in the chronic phase of the pilocarpine-SE model the TCA cycle is also impaired in the hippocampus. A loss of ^{13}C enrichment was also observed in both lactate (33%, $p<0.05$) and alanine (31%, $p<0.05$) in the liver of mice that developed SE, indicating a loss in glucose uptake or glycolytic activity in the liver. Chronic administration of F16BP restored the % 13C enrichment of all metabolites both peripherally and in the brain, except for lactate where a 16% decrease was observed in the hippocampus ($p<0.05$).
Overall this suggests that in the chronic epileptic hippocampal formation glucose metabolism is impaired by reductions in glucose uptake and activities of key TCA cycle enzymes. However, these impairments in glucose handling are not limited to the CNS, but also occur peripherally,

and the administration of F16BP restored both brain and peripheral glucose metabolism. We are currently investigating potential mechanisms for the improvement of glucose metabolism by F16BP.

Disclosures: T. McDonald: None. **M.P. Hodson:** None. **K. Borges:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.03/U11

Topic: C.01. Brain Wellness and Aging

Support: NIH RO1 NS070261

The Barrow Neurological Foundation

CIHR 1038081

Title: Sirtuins confer the neuroprotective activity of the ketogenic diet and the ketone body β-hydroxybutyrate

Authors: H. OH[1], S.-C. MA[1], A. J. SHACKELFORD[2,1], J. M. RHO[3], *D. KIM[1]
[1]Barrow Neurolog. Institute, St. Joseph's Hosp. & Med. Ctr., Phoenix, AZ; [2]Dept. of Dent. & Oral Hlth., A.T. Still Univ., Mesa, AZ; [3]Dept. of Pediatrics, Clin. Neuroscience, Physiol. and Pharmacol., Alberta Children's Hospital, Univ. of Calgary, Calgary, AB, Canada

Abstract: A growing number of studies indicate that metabolism-based treatments such as the high-fat anticonvulsant ketogenic diet (KD) and calorie restriction can exert neuroprotective activity, possibly through modulation of mitochondrial function and neuroinflammation. Although the specific mechanisms underlying these effects remain unclear, there are rapidly emerging links between cellular metabolism and nicotinamide adenine dinucleotide-dependent deacetylases (i.e., sirtuins, Sirt1-7), some of which affect epigenetics, neurometabolism, and inflammation. Given that administration of either the KD or β-hydroxybutyrate (βHB; a key metabolic byproduct of the KD) has been shown to attenuate neuronal death and pro-inflammatory responses against lipopolysaccharide (1μg/ml LPS; an endotoxin), a mitochondrial respiratory complex I inhibitor; 1 μM rotenone, and hydrogen peroxide (H_2O_2; 250 μM) (SFN 2016, #516.15), we investigated whether: 1) gene expression levels of Sirt1-7 are elevated in KD-fed mice brain and βHB-treated hippocampal HT22 cells or microglial BV2 cells; and 2) blockade of Sirt3 activity counteracts βHB-induced neuronal protection and anti-inflammatory effects. In hippocampus from C57BL/6J mice fed the KD for 2 wks, PCR analysis of genomic DNA using primers for Sirt1-7 revealed a significant increase in mRNA expression levels of all sirtuins compared with standard diet-fed mice. Consistent with these *in vivo* data, cell lines

exposed to 1 mM βHB for 24-hr led to upregulation of Sirt1-7 gene expression; notably, the most robust changes were seen in Sirt3 and Sirt6 expression. Pharmacological blockade of Sirt3 (1 ug/ml 5-amino-2-phenyl-benzoxazole) activity had no influence on HT22 cell viability, whereas βHB-induced neuronal protection against H_2O_2 was strongly reversed by inhibition of Sirt3 activity. Blocking Sirt3 activity in BV2 cells for 6-hr strongly enhanced release of pro-inflammatory mediators, compared to BV2 cells treated with either rotenone or LPS. βHB failed to induce anti-inflammatory effects under conditions of reduced Sirt3 activity. Collectively, our data support the notion that activation of sirtuins underlies some of the therapeutic benefits of βHB and the KD.

Disclosures: H. Oh: None. **S. Ma:** None. **A.J. Shackelford:** None. **J.M. Rho:** None. **D. Kim:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.04/U12

Topic: C.01. Brain Wellness and Aging

Support: Women Advancement Initiative, University of Hartford

College of Arts and Science, University of Hartford

NIH

Title: Ketogenic diet produces rapid, region-specific alterations in brain NAD^+/NADH

Authors: *M. ELAMIN[1], D. N. RUSKIN[2], S. A. MASINO[2], P. SACCHETTI[1]
[1]Neurosci. program, Dept. of Biol., Univ. of Hartford, Hartford, CT; [2]Neuroscience/Psychology, Trinity Col., Hartford, CT

Abstract: Numerous studies establish anti-epileptic effects of a ketogenic diet (KD), and recent research highlights its therapeutic potential in developmental disorders such as autism spectrum disorder and in neurodegenerative diseases such as Alzheimer's disease. A KD's composition of high fat, low carbohydrate, and moderate protein induces a metabolic shift that lowers glucose availability, increases ketone utilization as an energy source, and improves mitochondrial function. However fundamental mechanisms underlying its beneficial effects across diverse disorders are not well understood. Based on differential utilization of nicotinamide adenine dinucleotide (NAD) during glucose-based versus ketone-based generation of cellular ATP we hypothesized that a primary mechanism is altered levels of NAD. NAD is an essential metabolic coenzyme and a signaling molecule that exists in oxidized and reduced forms, NAD^+ and NADH, respectively. We fed normal rats ad libitum with regular chow or KD for either two days

or three weeks and quantified the regional NAD^+/NADH ratio in frontal cortex and hippocampus. In animals fed a KD for three weeks we observed region-specific effects: the NAD^+/NADH ratio was increased significantly in the hippocampus but not in the frontal cortex, thus suggesting that some brain regions may be more sensitive than others to this metabolic shift. Furthermore, the increased ratio in the hippocampus was detected already within two days of KD feeding and remained elevated, indicating an early as well as a persistent metabolic shift during diet administration. These effects paralleled metabolic changes in blood ketones: rats were also already in ketosis at two days of KD feeding. The hippocampus is a key brain region for seizure generation and memory formation and has a high metabolic rate - perhaps relevant to selective KD-induced changes in NAD^+/NADH. In general increasing NAD^+ is a coveted therapeutic endpoint - considered a marker for mitochondrial and cellular health, and postulated to increase longevity. Here we found that a KD induces a rapid and region-specific increase in NAD^+/NADH, a change that may be a primary mechanism behind diverse beneficial effects of this metabolic therapy.

Disclosures: **M. Elamin:** None. **D.N. Ruskin:** None. **S.A. Masino:** None. **P. Sacchetti:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.05/V1

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant AT008742

NIH Grant NS066392

NIH Grant NS065957

Title: Ketogenic diet shifts cerebrospinal fluid metabolome and has differential effects in responsive vs. non-responsive pediatric epilepsy patients

Authors: *D. N. RUSKIN[1], M. LINDEFELDT[2], N. FREEDGOOD[1], M. DAHLIN[2], S. A. MASINO[1]
[1]Trinity Col., Hartford, CT; [2]Karolinska Hosp., Stockholm, Sweden

Abstract: The low carbohydrate, high fat ketogenic diet (KD) can be an effective anticonvulsant treatment in some pharmacoresistant epileptic patients. Its mechanism(s) of action, however, remain uncertain. We performed metabolomic analysis of CSF samples taken before diet and during KD treatment in five complete responders and five complete non-responders to search for differential diet effects in these two groups. Responders and non-responders had similar age ranges and included both males and females. Seizure types and epileptic etiology or syndromes

varied widely, and did not appear to differ systematically between responders and non-responders. 271 metabolites were identified in CSF. Principal component analysis showed a strong effect of KD treatment on metabolites, particularly lipids, ketone bodies, carnitine derivatives, and sugars. Comparing responders and non-responders for the hallmark metabolic effects of KD, there was a greater elevation of ketone bodies and a larger drop in glucose in responders. Exploratory random forest analysis compared during-KD data between responders and non-responders to find other metabolites that could be relevant to differential seizure responsiveness. Top scoring metabolites included multiple lipids, amino acids, carbohydrates, and xenobiotics. These data show that KD feeding strikingly modifies the central metabolome, and that responders may have a stronger metabolic response to KD feeding.

Disclosures: D.N. Ruskin: None. **M. Lindefeldt:** None. **N. Freedgood:** None. **M. Dahlin:** None. **S.A. Masino:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.06/V2

Topic: C.01. Brain Wellness and Aging

Support: NINDS R21 NS098009

NINDS R01 NS076885

Title: Glycolytic inhibition with 2-deoxyglucose attenuates epileptiform activity following traumatic brain injury

Authors: *C. G. DULLA[1], D. CANTU[2], J. KOENIG[2]
[1]Neurosci., [2]Tufts Univ. Sch. of Med., Boston, MA

Abstract: Following a traumatic brain injury (TBI), post-traumatic epilepsy (PTE) can occur. Chronic seizures can be a significant cause of disability for TBI patients, especially when the seizures are refractory to medical anticonvulsant therapies. To study TBI, we have used a model known as controlled cortical impact (CCI) in mice. Using this approach we found cortical network hyperexcitability, increased glutamatergic signaling, and a loss of parvalbumin and somatostatin interneurons following injury. Glycolytic inhibition has begun to be explored as a therapeutic target for seizures, but has not yet been utilized to prevent post-traumatic epileptogenesis. We hypothesize that 2-deoxyglucose (2DG), a glucose analog that competitively inhibits glycolysis at the rate-limiting enzyme hexokinase, is neuroprotective and anti-epileptogenic following TBI. In vitro 2DG application attenuated epileptiform activity in acute cortical slices from injured brains. Additionally, in vivo 2DG treatment prevented the

development of epileptiform activity following injury and attenuated the loss of parvalbumin-expressing interneurons and the increase in reactive astrocytes following CCI. Our preliminary data also suggests that glycolytic inhibition with 2DG has different effects on the excitability of different neuronal subtypes, specifically that 2DG may decrease the excitability and membrane resistance of excitatory pyramidal cells while having no effect (or even increasing) the excitability of inhibitory interneurons. Our research supports a role for glycolytic inhibition in the prevention of epileptogenesis following TBI and may reveal a novel cell type-specific coupling of metabolism to neuronal excitability.

Disclosures: C.G. Dulla: None. **D. Cantu:** None. **J. Koenig:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.07/V3

Topic: C.01. Brain Wellness and Aging

Support: CONACYT FOSSIS 262010

DGAPA-UNAM to LARC

Title: Correlation between tryptophan catabolism and cognitive decline in old women

Authors: *L. A. RAMOS[1,2], P. CARRILLO-MORA[3], B. GARCÍA[3], D. GONZÁLEZ-ESQUIVEL[4], D. RAMÍREZ-ORTEGA[4], C. RIOS[4], V. PÉREZ-DE LA CRUZ[4], B. PINEDA[5], G. ROLDÁN-ROLDÁN[2]
[1]Univ. Nacional Autónoma de México, Ciudad DE Mexico, Mexico; [2]Lab. de Neurobiología de la Conducta, Dept. de Fisiología, Facultad de Medicina, Univ. Nacional Autónoma de México, México, México, México city, Mexico; [3]Dept. de Neurorehabilitación, Inst. Nacional de Rehabilitación, México, México city, Mexico; [4]Dept. de Neuroquímica, [5]Lab. de Neuroinmunología, Inst. Nacional de Neurología y Neurocirugía Manuel Velasco Suárez, S.S.A., México, México city, Mexico

Abstract: Aging is a multifactorial degenerative process and is characterized by progressive deterioration in physiological functions and metabolic processes; these changes drive to numerous age-related disorders. Cellular alterations during aging process involve oxidative stress, inflammation, mitochondrial dysfunction, cognitive and immune response decline. Recently, it was found that changes modifications in the tryptophan metabolism are also observed during the aging. Tryptophan is an essential amino acid required for synthesis of proteins and it is mainly metabolized via the kynurenine pathway conducing to several metabolites with neuroactive and/or redox properties. Particularly, kynurenic acid -an

endogenous antagonist of α7nACh and NMDA receptors- and quinolinic acid, an agonist of NMDA receptors, have been related with neurodegenerative diseases. Due to kynurenine pathway metabolites have been related with aging and some ageing-related diseases, we investigated whether the imbalance of kynurenine pathway components could be related with cognitive decline and alteration in redox status in old women. Participants ranged in age from 50 to 95 years. A brief neuropsychological tests serie (NEUROPSI), standardized, was performed. The NEUROPSI tests serie includes assessment of orientation, attention, memory, language, visuoperceptual abilities, motor skills, and executive functions. Serum levels of tryptophan, kynurenine, kynurenic acid and QUIN were determined in seventy old women without dementia, as well as lipoperoxidation and GSH levels in the same samples. Cognitive decline increased during aging. GSH levels decreased whereas lipid peroxidation increased during aging. Tryptophan levels decreased in relation to the age and correlated with the cognitive decline ($P < 0.0019$). Additionally, QUIN / Trp ratio also correlated with cognitive decline. Our data suggest that tryptophan catabolism is more active as the age advances and it is involved in the cognitive detriment found in older women.

Disclosures: **L.A. Ramos:** None. **P. Carrillo-Mora:** None. **B. García:** None. **D. González-Esquivel:** None. **D. Ramírez-Ortega:** None. **C. Rios:** None. **V. Pérez-de la Cruz:** None. **B. Pineda:** None. **G. Roldán-Roldán:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.08/V4

Topic: C.10.Tauopathies, Tau-dementias, and Prion diseases

Support: NIH Grant R01GM088801

NIH Grant R01AG041274

NIH Grant R01HD 086977

Title: Inhalation anesthetics induce abnormal locomotor activity in mice

Authors: *H. TON[1], L. YANG[2], Z. XIE[3]
[1]Dept. of Anesthesia, Critical Care and Pain Med., Massachusetts Gen. Hosp., Boston, MA; [2]Massachusetts Gen. Hospital- Harvard Med. Sch., Boston, MA; [3]Anesthesia, Critical Care and Pain Med., Massachusetts Gen. Hosp. and Harvard Med. Sch., Charlestown, MA

Abstract: Delirium is one of the most common postoperative complications in geriaric patients and can lead to 2- to 20-fold increase in mortality, long term functional impairment, postoperative cognitive dysfunction, and increased costs of medical care. However, its causes,

neuropathogenesis and targeted intervention(s) remain largely to be determined, partially owing to the lack of animal model(s). The mouse activity was recorded before, during and after anesthesia with sevoflurane or isoflurane [0.2-, 0.5-, 1- and 2-fold minimum alveolar concentration (MAC)] at 37^0 C. The total walking distance, velocity of movement and the time visiting the central zone of the box were measured and quantified as indexes of hyperactivity and agitaiton-like behaviors. Both isoflurane and sevoflurane at all the tested concentrations changed the motility of the mice in a time-dependent manner. During the anesthesia, sevoflurane and isoflurane markedly increased locomotion. After anesthesia, the locomotion increased in a very short time (within 5 min) and markedly reduced up to 2 hours after treatment. Interestingly, food intake and body weight decrease up to 48 hours after the anesthesia in the anesthetized mice as compared to control mice. Our results provide the evidence for the first time that inhalation anesthetics induce short-term hyperactivity and agitation-like behavior, and long-term depressive-like behavior in the mice. These data would help us to establish an animal model to study emergence and postoperative delirium, ultimately leading to the understanding the neuropathogenesis of postoperative delirium and the development of targeted inverventions.

Disclosures: H. Ton: None. **L. Yang:** None. **Z. Xie:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.09/V5

Topic: C.01. Brain Wellness and Aging

Support: Indian Council of Medical Research

International Brain Research Organisation

Title: Altered levels of neurotrophic factors and neurotransmitters in the brain as the probable causes of decreased longevity of WNIN obese rats

Authors: *S. GHOSH[1], J. K. SINHA[1,2], N. GIRIDHARAN[1], M. RAGHUNATH[1]
[1]NATIONAL INSTITUTE OF NUTRITION, Hyderabad, India; [2]CSIR-CENTRE FOR CELLULAR AND MOLECULAR BIOLOGY, Hyderabad, India

Abstract: Wistar NIN obese (WNIN/Ob) rats developed at the National Institute of Nutrition are the heaviest inbred rat strain in the world. These rats are hyperphagic, hyperinsulinemic, hyperleptinemic and have reduced longevity (an average lifespan of 15-18 months in contrast to 36 months in normal Wistar rats). In the WNIN/Ob rats, we intend to delineate the factors responsible for reduced longevity. Neurotrophic factors are responsible for the survival of developing neurons and the maintenance of mature neurons. We have estimated levels of key

neurotrophic factors using BioPlex assay and done neuro-glial profiling using Immunohistochemistry in different brain regions of these rats (n=6). As Glutamate (Glu) and Gamma-aminobutyric acid (GABA) are the major excitatory and inhibitory neurotransmitters in mammalian CNS, we have looked at their levels in different brain regions of WNIN/Ob rats (n=4) and age-matched normal rats (n=4) using Magnetic Resonance Spectroscopy. We have evaluated if there are any volumetric differences in the brain of WNIN/Ob rats in contrast to the age-matched controls using Magnetic Resonance Imaging. Our findings show that the levels of key neurotrophic factors like BDNF and IGF-1 are altered in the WNIN/Ob rats. MRS data indicates altered neurotransmitter metabolism in the brain of WNIN/Ob rats. But there are no significant volumetric changes in the brain of the WNIN/Ob rats when compared to controls. Altered levels of neurotrophic factors and neurotransmitters in the brain are one of the many factors behind decreased longevity of WNIN obese rats.

Disclosures: S. Ghosh: None. **J.K. Sinha:** None. **N. Giridharan:** None. **M. Raghunath:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.10/V6

Topic: C.01. Brain Wellness and Aging

Support: NRF

Title: Adiponectin receptor 1 regulates AKT-FOXO mediated cell survival via facilitating insulin signaling

Authors: *M.-W. KIM, G.-H. YOON, M.-G. JO, F. U. AMIN, M. IDREES, M.-O. KIM
Gyeongsang Natl. Univ., Jinju-Si, Gyeongsangnam-Do, Korea, Republic of

Abstract: Adiponectin, adipocyte-derived hormone, is considered as master regulator for anti-diabetes and insulin resistance by sensitizing insulin signaling on peripheral tissues. Since neuro-pathology has been termed as Type-3-diabetes, adiponectin-adiponectin receptors have been regarded as a therapeutic target. Previously, we reported that AdipoR1 deficiency exhibited neuronal apoptosis and spatial learning and memory impairment. To extend the mechanisms of how AdipoR1 regulates neuronal apoptosis, we focused on the insulin signaling-apoptosis axis. Using PEI-mediated gene therapy for in vivo knockdown, we found that AKT phosphorylation at serine 473 region (p-AKT) was highly down-regulated in AdipoR1 shRNA group. Accordingly, AKT-mediated glucose uptake by insulin-stimulation was decreased. Moreover, we checked whether down-regulated p-AKT by AdipoR1 suppression attenuates FOXO1 phosphorylation. We confirmed this AKT mediated FOXO1 phosphorylation was prevented by AdipoR1 shRNA.

This facilitates FOXO1 nuclear translocation as well as transcribing its apoptotic-related genes. Taken together, our findings suggest a specific role of AdipoR1 in insulin signaling mediated cell survival, and point to the modulation of AdipoR1 as a therapeutic target of neurodegeneration.

Disclosures: M. Kim: None. **G. Yoon:** None. **M. Jo:** None. **F.U. Amin:** None. **M. Idrees:** None. **M. Kim:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.11/V7

Topic: C.01. Brain Wellness and Aging

Support: NIH RO1 Grant RN0136

Title: Oxidative stress: Possible modulator of hormone replacement therapy action

Authors: *P. DUONG[1], C. MCCUISTON[2], R. CUNNINGHAM[1]
[1]CANDR, [2]Texas Col. of Osteo. Med., UNT Hlth. Sci. Ctr., Fort Worth, TX

Abstract: One of the hallmarks that delineates reproductive aging in women is menopause. Menopause is a gradual process that begins to manifest in women as they approach 45-55 years of age. Prior studies have found equivocal effects of hormone replacement therapy (HRT) on brain health for women in menopause. HRT initiation prior to the manifestation of menopause has been shown to be protective. However, HRT initiation in women 10 years post-menopausal can induce adverse effects, such as stroke. Oxidative stress is a key characteristic in aging, menopause, and stroke. Therefore, we propose that oxidative stress may be a switch that determines if HRT is neuroprotective or neurotoxic. To examine our hypothesis we used an immortalized neuronal cell line $1RB3AN_{27}$ (N27) derived from female fetal mesencephalic tissue. Hydrogen peroxide (H_2O_2) was used as an oxidative stressor to model menopause. Common HRTs prescribed by physicians include testosterone (T), 17β-estradiol (E2), and membrane impermeable - dihydrotestosterone (DHT-BSA). Therefore, we exposed N27 cells to HRT either before or after H_2O_2 treatment. Cell viability was assessed using MTT. Results showed that HRT prior to H_2O_2 was protective, regardless of the type of HRT. However, if HRT was applied after H_2O_2, N27 cell loss was exacerbated. These results indicate that oxidative stress may be a switch that determines if HRT is neuroprotective or neurotoxic. Furthermore, this study provides further support and extends the Healthy Cell Bias of Estrogen Signaling.

Disclosures: P. Duong: None. **C. McCuiston:** None. **R. Cunningham:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.12/V8

Topic: C.01. Brain Wellness and Aging

Title: Curcuma treatment prevents alteration ofneuronal morphology in the limbic system of aging rats

Authors: *R. A. VAZQUEZ, SR[1], B. VIDAL[2], B. FLORAN[5], D. GNECCO[3], G. FLORES[4]
[1]Inst. De Fisiología Benemérita Univ. Autónoma De Puebla, Puebla, Mexico; [2]Inst. de Fisiologia, Benemerita Univ. Autonoma de Puebla, Puebla, Puebla, Mexico; [3]Ctr. de quimica, Inst. de Ciencias, [4]Inst. de Fisiologia, Benemerita Univ. Autonoma de Puebla, Puebla, Mexico; [5]Dept. de Fisiología Biofícia y Neurociencias, Ctr. de investigaciones y estudios avanzados IPN, Mexico City, Mexico

Abstract: Curcuma is a natural compound that has shown neuroprotective properties, and has been reported to prevent aging and improve memory. While the mechanisms underlying these effects are unclear, they may be related to increases in neural plasticity. Morphological changes have beenreported in neuronal dendrites in the limbic system in animals and elderly humans with cognitive impairment. In this regard, there is a need to use alternative therapies that delay the onset of morphologies and behavioral characteristics of aging. Therefore, the objective of this study was toevaluate the effect of curcuma on cognitive processes and dendritic morphology of neurons in the prefrontal cortex , the CA1 and CA3 regions of the dorsal hippocampus, the dentate gyrus, and the basolateral amygdala of aged rats. 18-month-old rats were administered curcuma 100 mg/kg daily for 60 days. After treatment, recognition memory was assessed using the novel object recognition test. Curcuma-treated rats showed a significant increase in the exploration quotient. Dendritic morphology was assessed by Golgi-Cox staining and followed by Sholl analysis. Curcuma-treated rats showed a significant increase in dendritic spine density and dendritic length in pyramidal neurons of the PFC, the CA1 and CA3, and the BLA. The preservation of dendriticmorphology was positively correlated with cognitive improvements. Our results suggest that curcuma induces modification of dendritic morphology in the aforementioned regions. These changes may explain how curcuma slows the aging process that has already begun in these animals, preventing deterioration in neuronal morphology of the limbic system and recognition memory.

Disclosures: R.A. Vazquez: None. **B. Vidal:** None. **B. Floran:** None. **D. Gnecco:** None. **G. Flores:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.13/V9

Topic: C.01. Brain Wellness and Aging

Support: NIH R01 grant

NIH R56 grant

Donation from RoseMary Kubik

Title: Age-dependent vulnerability of axons to aglycemia is not caused by a decline in the astrocyte-neuronal lactate shuttle

Authors: *C. BASTIAN, C. DOHERTY, C. FRANKE, A. FARIS, S. BRUNET, S. BALTAN
Cleveland Clin. Fndn., Cleveland, OH

Abstract: The effects of aging on the central nervous system (CNS) predominantly impact the white matter (WM) compared to the gray matter. Age-dependent WM changes contribute to many CNS neurodegenerative diseases including dementia, ataxia, motor neuron diseases and stroke. We have previously shown that aging axon function is more vulnerable to ischemia and that age-related structural changes in myelin, mitochondria, and endoplasmic reticulum are associated with reduced energy production and Ca^{2+} dysregulation. Because it is well-established that glial cells support axon function, we tested the impact of aging on glial cells and its impact on axon-glia support. The astrocyte-neuron lactate shuttle (ANLS) is required to support axon function by providing lactate during periods of aglycemia or increased activity as an alternative energy substrate to glucose. We hypothesized that aging axon function is more vulnerable to reduced glucose availability and that this vulnerability is partly mediated by a decline in the ability of aging astrocytes to provide lactate to axons.
We utilized, mouse optic nerve (MON), a pure WM tract obtained from 2- or 12-month old mice to quantify axon function using electrophysiology. Axon function was quantified by monitoring the area under evoked compound action potentials (CAPs). To assess structural changes in aging astrocytes, MONs were immunostained with GFAP (astrocytes) and Sytox (nuclei). Aging astrocytes displayed thicker processes that extended parallel to axons. This contrasted with young astrocytes, which had shorter, thinner processes that traversed axons vertically. Aging axons were more susceptible to aglycemic injury (0 mM glucose for 1 h), recovering significantly less compared to young axons. However, when incubated with high glucose (30 mM for 1h) to increase glycogen stores in astrocytes to provide lactate during aglycemia, both young and aging axons showed delayed loss-of-function during aglycemia and recovered more following aglycemia. Furthermore, aging axon function recovery benefited more compared to

young axons when L-lactate was supplied during aglycemia. However administration of L-lactate following aglycemia did not improve axon function recovery in either young or aging axons.
Overall our results suggest that aging axons are more vulnerable to aglycemia and there are age-dependent structural changes in astrocytes. However, ANLS function to support axon function is not the underlying reason for this increased vulnerability. Further studies are in progress to uncover the mechanisms of increased vulnerability of aging axons to lack of glucose.

Disclosures: C. Bastian: None. **C. Doherty:** None. **C. Franke:** None. **A. Faris:** None. **S. Brunet:** None. **S. Baltan:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.14/V10

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant NS050396

NIH Grant RO1ES022936

AHA 15PRE25100000

AHA 14PRE2003500007

Dan Marino Foundation

VISRA Program Scholarship

Title: C-terminus of HSC70-Interacting Protein (CHIP) is a functional homologue of Atg5

Authors: *B. N. LIZAMA-MANIBUSAN[1], A. M. PALUBINSKY[2], J. W. MCLAUGHLIN[3], D. D. SZYMKIEWICZ[3], V. A. RAVEENDRAN[3], A. M. MOORE[3], B. MCLAUGHLIN[4]
[2]Neurosci. Grad. Program, [1]Vanderbilt Univ. Med. Ctr., Nashville, TN; [4]Neurol., [3]Vanderbilt Univ., Nashville, TN

Abstract: Autophagic protein and organelle degradation is an essential, highly conserved process that regulates normal and pathophysiological processes such as cancer and neurodegeneration. Autophagy Protein 5 (Atg5) and Atg12 work cooperatively in the formation of pre-autophagosomes, and mutations in Atg5 have been shown to result in profound deficits in autophagic signaling. Mice with global deletion of Atg5 die shortly after birth and Atg5 mutations were recently identified in two patients with ataxia. This phenotype is consistent with that of patients with an early onset, recessive form of spinocerebellar ataxia who have mutations

in the E3-ligase CHIP.
Animals with CHIP deficiency exhibit profound deficits in motor and cardiovascular function and premature death. Our prior proteomic and biochemical analysis of CHIP knockout (KO) animals indicates that CHIP is essential for energetic and redox homeostasis. We found that in response to acute bioenergetic failure, CHIP relocates to mitochondria and colocalizes with the autophagy protein LC3. In this work, we used electron and fluorescence microscopy to demonstrate that CHIP KO mouse embryonic fibroblasts (MEFs) exhibit increased numbers of swollen, misshapen mitochondria with malformed cristae. Similarly, Atg5 KO cells demonstrate mitochondria with unstructured or inapparent cristae.
To assess the CHIP domains necessary for mitochondrial targeting, we transfected CHIP KO primary neurons with plasmids containing the human CHIP sequence (hCHIP) or mutated CHIP sequences in which E3 ligase activity is eliminated (H260Q) or TPR-domain binding is blocked (K30A). We found that these mutations did not affect the ability of CHIP to localize to mitochondria; however, they did abolish the ability of CHIP-tagged mitochondria to undergo mitophagy. We also examined the ability of Atg5 to compensate for defects in mitophagy caused by CHIP deletion by transfecting CHIP KO MEFs and primary neurons with an Atg5 overexpression plasmid. Given the rapid localization of CHIP to mitochondria during stress and the role of CHIP in maintaining basal energetic homeostasis, we propose that CHIP is essential for mitophagy in the neuronal autophagic pathway. Ongoing work is aimed at identifying the interactions of CHIP with autophagy proteins and in non-mitophaghic pathways, as well the overall role of CHIP/Atg signaling in response to bioenergetic stress.

Disclosures: B.N. Lizama-Manibusan: None. **A.M. Palubinsky:** None. **J.W. McLaughlin:** None. **D.D. Szymkiewicz:** None. **V.A. Raveendran:** None. **A.M. Moore:** None. **B. McLaughlin:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.15/V11

Topic: C.01. Brain Wellness and Aging

Support: 5P20GM109025

Title: High-fat diet-induced insulin disruption in CX3CR1 knockout mice on dementia-related pathology

Authors: *A. S. MURTISHAW[1], M. M. BOLTON[2], A. J. BOREN[2], A. M. SALAZAR[2], J. E. TOUGHLIAN[2], A. A. ORTIZ[2], J. W. KINNEY[2]
[1]Neurosci., UNLV Neurosci. Doctoral Student, Las Vegas, NV; [2]Psychology, Univ. of Nevada Las Vegas Dept. of Psychology, Las Vegas, NV

Abstract: Alzheimer's disease (AD) is the most common form of dementia. Inflammation and insulin signaling perturbations have emerged as important risk factors associated with AD. Recent studies have also demonstrated an interaction between alterations in insulin signaling and inflammation. Many studies support the relationship between AD and metabolic disorders, particularly diabetes mellitus. Fractalkine (CX3CL1) is a chemokine that has been shown to protect the brain from the damaging consequences of chronic inflammation by interacting with its obligate receptor (CX3CR1) on microglia. Additionally, recent studies have demonstrated that the CX3CL1/CX3CR1 system plays a regulatory role in pancreatic β-cell function and insulin secretion. To further explore the relationship that fractalkine plays with insulin signaling and dementia-related pathology, we administered a high-fat diet (HFD) to CX3CR1 knockout mice for six months to evaluate alterations in dementia-related pathologies. We chose HFD as our diabetic model to more closely mirror our clinical population of insulin resistance stemming from obesity and Type-2 diabetes. Animals were tested in the novel object recognition task and spatial learning in the Barnes maze. Following completion of behavioral training, brains were removed for biochemical examination, including markers of inflammation, metabolic alterations, and histopathological markers relevant to dementia.

Disclosures: A.S. Murtishaw: None. **M.M. Bolton:** None. **A.J. Boren:** None. **A.M. Salazar:** None. **J.E. Toughlian:** None. **A.A. Ortiz:** None. **J.W. Kinney:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.16/V12

Topic: C.01. Brain Wellness and Aging

Support: Department of Science and Technology EMR/2015/002178/HS

ICMR

DBT

UGC UPE

Title: HDAC2 and associated co-repressor sin3a regulate synaptic plasticity gene expression and memory consolidation during scopolamine-induced amnesia

Authors: *S. SRIVAS, M. K. THAKUR
Inst. of Science, BHU, Varanasi, India

Abstract: Memory is a vital brain function which declines in several conditions including physiological state of advancing age and in neurodegenerative pathologies. Memory decline, also

referred as amnesia, may affect different stages of memory processes. Memory consolidation is a protein synthesis dependent process. A group of synaptic plasticity genes, neuronal immediate early genes (IEGs), play a crucial role in memory consolidation. The expression of these genes changes during physiological and pathological conditions. Recent reports have shown that expression of synaptic plasticity genes is modulated by chromatin remodeling due to DNA methylation and histone post-translational modification, which in turn regulate learning and memory. However, the epigenetic regulation of IEGs is largely unexplored in amnesia. Therefore, the present study elucidates the expression and epigenetic regulation of IEGs (Arc, Egr1, Homer1 and Narp) in a male *Mus musculus* model of amnesia by administration of scopolamine. Scopolamine-induced amnesia was validated by radial arm maze test followed by expression analysis of neuronal IEGs. The expression of chromatin modifying enzymes (CREB binding protein (CBP), histone deacetylase (HDAC) 2, DNA methyltransferase (DNMT) 1, 3a and 3b) was studied at mRNA and protein level. DNA methylation and histone acetylation (H3K9/14Ac) status was assessed by methylated DNA immunoprecipitation and chromatin immunoprecipitation followed by qPCR at the promoter of neuronal IEGs. The results revealed that scopolamine significantly reduced memory consolidation and expression of neuronal IEGs, but increased the expression of DNMT1 and HDAC2 in the hippocampus. Further, scopolamine treatment increased DNA methylation and decreased histone H3K9/14 acetylation at the promoter of IEGs. As, HDAC2 binds on DNA in conjunction with DNMT associated co-repressor complex, we assessed the expression of co-repressor molecule Sin3a at mRNA and protein level in the hippocampus of mice. We found increase in the expression of Sin3a during amnesia. Further, we silenced Sin3a in the hippocampus of amnesic mice by administration of siRNA. Sin3a silencing in amnesic mice increased memory consolidation by increasing the expression of neuronal IEGs. Taken together, our study showed that DNMT1 and HDAC2 expression is associated with downregulation of IEGs in the hippocampus and thereby decline in memory consolidation during amnesia. Further, Sin3a inhibition restored memory consolidation in amnesic mice. Such alteration in HDAC2 and associated Sin3a complex may be used as a target to develop potential therapeutic interventions for amnesia.

Disclosures: S. Srivas: None. **M.K. Thakur:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.17/V13

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant AG047652

Title: Optogenetic function is maintained through late aging (25 mo) in neurons from ChR2-eyfp (vGlut2 and vGAT) BAC mice

Authors: *D. W. DUBOIS, K. S. MONTGOMERY, A. S. FINCHER, E. A. BANCROFT, V. E. PROVASEK, E. A. MIGUT, D. A. MURCHISON, W. H. GRIFFITH
Dept. of Neurosci. and Exptl. Therapeut., Texas A&M Univ. Hlth. Sci. Ctr., Bryan, TX

Abstract: Our lab studies the relationship between altered synaptic physiology, calcium homeostasis and age-related cognitive impairment in the rodent basal forebrain (BF). Our goal is to understand how youthful synapses can be preserved and cognitive decline prevented during aging. The current study is designed to support the use of optogenetic (vGlut2- and vGAT-ChR2 (H134R)-eYFP BAC) mice as aging models. These strains express the light-sensitive channelrhodopsin 2 (ChR2) in neurons with vesicular transporters specific to glutamatergic and GABAergic neurons respectively. We utilize patch-clamp recording, 470nm light stimulation, fura-2 microfluorimetry and behavioral testing in young (2-8 mo), middle (10-14 mo) and aged (17-25 mo) mice. We have found that ChR2 expression is functionally maintained during aging. In GABAergic BF neurons, light-evoked ChR2 inward currents (peak current density, pA/pF) were unchanged with age (young: 4.6 ± 1.42, middle: 3.18 ± 0.59, aged: 4.23 ± 1.41; n = 6-11). ChR2 current densities were also maintained in glutamatergic thalamic neurons during aging (young: 4.61 ± 1.42, middle: 3.17 ± 0.59, aged: 4.23 ± 0.58; n = 9-14). We next sought to determine whether our optogenetic mouse model displayed age-related changes in synaptic transmission, calcium buffering and behavioral status as observed in our rat BF model. Optogenetic inhibitory postsynaptic currents (oIPSCs) were evoked using brief light pulses (2-5 ms) in BF slices of vGAT-ChR2 mice. There was a decrease in oIPSCs with age: young, 37.5 ± 5.1 pA/pF, n = 40; aged, 22.3 ± 4.7 pA/pF, n=17 (p = 0.058). There was also an increase in intracellular calcium buffering during aging. A significant decrease in slope (increased buffering) of buffering curves (Ca-transient amplitude x stimulus duration) was seen in BF neurons from aged mice of both genders with no difference between genotypes. This parallels previous findings in rat BF. The slope values for WT mice were: young, 46.8 ± 4.8 (n=26) and aged, 31.3 ± 3.3 (n=40, $p<0.01$). There were no differences in baseline [Ca] in any of the groups. Neurons were confirmed as cholinergic by spot-checking with scRT-PCR. In 16/16 such neurons ChAT sequence was detected. We assessed behavioral status in mice using the Barnes Maze for a spatial memory task, open field for a measure of anxiety and the rotarod to monitor motor coordination. Preliminary data, as predicted, showed age-related deficits across the battery of tests. We are currently refining our analysis of behavioral differences across genotypes, genders and age. These results demonstrate that these optogenetic BAC mice are excellent models for investigation of synaptic aging.

Disclosures: D.W. Dubois: None. **K.S. Montgomery:** None. **A.S. Fincher:** None. **E.A. Bancroft:** None. **V.E. Provasek:** None. **E.A. Migut:** None. **D.A. Murchison:** None. **W.H. Griffith:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.18/V14

Topic: C.01. Brain Wellness and Aging

Support: NIA Grant 1R15AG050292

Alzheimer's Association Grant NPSPAD 247219

Title: Utilization of KKA_y diabetic model to elucidate the role of metabolic dysfunction in the development of Alzheimer's disease pathology

Authors: *J. GRIZZANTI[1], S. SAHU[2], P. RAMAN[2], H.-G. LEE[3], G. CASADESUS[1]
[1]Sch. of Biomed. Sci., Kent State Univ., Kent, OH; [2]Northeast Ohio Med. Univ., Rootstown, OH; [3]Univ. of Texas at San Antonio, San Antonio, TX

Abstract: A strong body of evidence exists suggesting that type II diabetes and obesity are risk factors for the development of Alzheimer's disease. Furthermore, numerous studies have utilized diet to induce obesity in rodent models of AD and have demonstrated increases in AD pathology related markers. However, few studies have utilized obese and/or diabetic models to assess the effects of metabolic hormone resistance on hallmark AD pathology in rodent models lacking AD-related genes. Here, we utilized KKAγ mice, a model that demonstrates a robust diabetic and obese phenotype, to survey the effects of insulin and leptin resistance on markers that contribute to the production of AD hallmark pathology. KKAγ mice showed significantly elevated body weight, blood glucose levels, free fatty acids, and blood leptin levels compared to littermate controls. Interestingly, KKAγ mice showed no significant changes in AD pathology-associated markers within the brain 4 months post development of the diabetic phenotype. Together, these data suggest that the ability of metabolic dysregulation to induce AD pathology may be mediated via fundamental neurotoxic mechanisms such us inflammation, mitochondrial dysregulation, and/or increased oxidative stress rather than directly through altering pathology related aspects.

Disclosures: J. Grizzanti: None. **S. Sahu:** None. **P. Raman:** None. **H. Lee:** None. **G. Casadesus:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.19/V15

Topic: C.01. Brain Wellness and Aging

Support: CFAR Grant P30 AI036214.

Title: The influence of childhood trauma, major depressive disorder and telomere length on HIV-associated neurocognitive disorders

Authors: *J. S. WOMERSLEY, G. SPIES, G. TROMP, S. HEMMINGS, S. SEEDAT
Univ. of Stellenbosch, Cape Town, South Africa

Abstract: Background: HIV-associated neurocognitive disorders (HAND) continue to prevail in countries affected by HIV/AIDS, despite improved access to antiretroviral therapies. Previous research has suggested that this is in part due to the complex interactions between stress-related exposures and depression, which may aggravate the development of neurocognitive impairment. Telomere length (TL) attrition is a marker of biological aging that has been independently associated with childhood trauma (CT), depression and HAND. We therefore sought to investigate whether TL shortening may act as a biomarker for HAND when examined in the context of CT and depression. **Methods:** HIV-positive (n=133) and negative women (n=150) underwent a battery of neuropsychological tests to measure seven domains of cognitive function: motor skill, verbal fluency, attention and working memory, processing speed, learning, recall, and executive function, from which a global cognitive score was calculated. Participants also completed the Childhood Trauma Questionnaire and the Centre for Epidemiological Studies Depression Scale. Quantitative polymerase chain reaction using primers specific to telomeric repeats and the reference gene human β-globin was performed on DNA extracted from peripheral blood mononuclear cells. Baseline differences in relative TL and neuropsychological parameters between HIV-positive and negative individuals were assessed using a t-test. The relationship between TL, cognitive function, CT and depression was probed using multiple linear regression models with age as a covariate. Data were analysed using the R statistical language, and an alpha value of less than 0.05 was deemed statistically significant. **Results:** HIV-positive individuals had significantly reduced relative TL ($t(275.45) = 5.731$, $p<0.0001$) and self-reported higher levels of CT ($t(262.59) = -7.671$, $p<0.0001$). Regression analysis revealed that including the interaction between CT and HIV status in the statistical model explained significantly more of the variance in TL ($p=0.035$). HIV status, CT, depression and TL individually did not affect cognitive function. However, the interaction of CT and depression explained significantly more of the variance in global cognitive scores across participants ($p=0.022$). In contrast, including the interaction between depression and TL explained significantly more of the variance in global

cognitive function in HIV positive individuals (p=0.025). **Conclusions:** Our data suggest that HIV is associated with decreased TL and that the interaction between this biomarker and psychological ill health may influence cognitive status.

Disclosures: J.S. Womersley: None. **G. Spies:** None. **G. Tromp:** None. **S. Hemmings:** None. **S. Seedat:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.20/V16

Topic: C.01. Brain Wellness and Aging

Support: FONDECYT 11160651

FONDECYT 1160724

CONICYT- PFB 12/2007

Title: Regulation of glucose metabolism in neurons by Wnt signaling and a possible molecular link with the adipokines in the context of Alzheimer disease and obesity interaction

Authors: *P. CISTERNAS[1], N. C. INESTROSA[2]
[1]CARE-UC, Pontificia Univ. Católica de Chile, Santiago, Chile; [2]Ctr. For Aging and Regeneration (CARE),P. Catholic Univ. of Chile, Santiago, Chile

Abstract: The brain is an organ with a high rate of glucose consumption and in several disorders it has been described a decrease in the brain capacity to utilize glucose including in Alzheimer´s disease (AD) and obesity. Despite the importance of this process little is know about the regulation in this process in both pathologies. In the lasts year has been described that obesity (increase in fat tissue) increase the risk of AD in several models. The proposal mechanism for explain this results evolve the action of adipokines, released by the fat tissue. In the present work we described for first time the effect of Wnt signaling activation over the glucose metabolism. Also, we described in cortical neurons that the effect of Wnt ligands are similar with the effects of adiponectin (an adipokine down regulated during obesity) over the glucose metabolism. In both cases we used glucose radiolabeled analogous to determine the values of K_m and V_{max} in primary cortical neurons and we observed an increase in the affinity for glucose uptake in both cases. Also, we observed different changes in the activity of pentose phosphate pathway (PPP) depended of the Wnt ligands used. This study suggests that the Wnt and adiponectin signaling stimulates the energy metabolism of neurons, and that these effects could be central to prevent metabolic impairment described in obesity and AD.
Supported by CONICYT- PFB 12/2007 to N.C.I., FONDECYT (no. 1120156) to N.C.I., and

FONDECYT (no. 11160651) to P.C. We also thank our special grants “The role of K^+ on Hypertension and Cognition” from the Sociedad Química y Minera de Chile (SQM).

Disclosures: P. Cisternas: None. **N.C. Inestrosa:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.21/V17

Topic: C.01. Brain Wellness and Aging

Support: National Research Foundation of Korea(NRF) Grant 2012R1A6A1028677

National Research Foundation of Korea(NRF) Grant 2015R1D1A1A01058350

Title: Effect of phycoerythrin-derived tryptic peptide of Pyropia yezoensis on glutamate-induced endoplasmic reticulum stress and neuronal senescence

Authors: *J. OH
Pukyong Natl. Univ., Busan, Korea, Republic of

Abstract: Glutamate recycling in synaptic cleft is impaired by increasing age, which induced glutamate accumulation that over-stimulates post-synaptic neurons. Glutamate-induced excitotoxicity has been involved in various neurodegenerative disorders such as cognitive dysfunction. Previous studies have demonstrated that peptides from the edible red alga *Pyropia yezoensis* have antioxidant and chemoprotective activities in various cell lines. This study was to investigate whether a phycoerythrin-derived tryptic peptide of *Pyropia yezoensis* (PYP) reduces glutamate-induced excitotoxicity and neuronal senescence in primary rat hippocampal neurons. Exposure to 100 μM glutamate significantly decreased cell viability and increased expression of endoplasmic reticulum (ER) stress response protein, glucose-regulated protein 78 (GRP78) from 60 min after glutamate exposure, which were downregulated by pretreatment of PYP (1 μg/ml). The glutamate-induced increase in GRP78 expression was downregulated by blockade of NMDA receptor and inhibition of JNK phosphorylation, MK801 (10 μM) and SP600125 (10 μM), respectively, and also phosphorylation of JNK by glutamate was decreased by blockade of NMDA receptor. Pretreatment of PYP downregulated glutamate-induced increase in GRP78 expression and JNK phosphorylation, which was abolished by inhibitions of TrkB receptor, PI3K, and ERK1/2, with cyclotraxin B (200 nM), LY294002 (20 μM), and SL327 (10 μM), respectively. In addition, PYP downregulated increase in GRP78 expression, senescence-associated β-galactosidase activity and neurite degeneration in aging hippocampal neurons. These findings indicate that activation of TrkB receptor-mediated ERK1/2 by PYP attenuates glutamate-induced ER stress and could contribute to survival of hippocampal neurons with age.

Disclosures: **J. Oh:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.22/V18

Topic: C.01. Brain Wellness and Aging

Support: UJAT-IB-2015-04 PFI2015

Title: Determination of Cholesterol 24 Hydroxylase (CYP46A1) in hippocampus of rats with obesity induced by different hypercaloric diets

Authors: *N. GOMEZ-CRISOSTOMO[1], E. MARTÍNEZ-ABUNDIS[2], E. DE LA CRUZ-HERNÁNDEZ[2]
[1]UNIVERSIDAD JUAREZ AUTONOMA DE TABASCO (DIVISION A, Comalcalco, Mexico; [2]LABORATORIO DE INVESTIGACION EN ENFERMEDADES METABÓLICAS E INFECCIOSAS, UNIVERSIDAD JUAREZ AUTONOMA DE TABASCO (DIVISION A, COMALCALCO, TABASCO, Mexico

Abstract: Several studies have described an association between neurodegenerative disorders and abnormalities in the metabolism of cholesterol, related with the obesity and metabolic syndrome. The cholesterol in the Central Nervous System (CNS) is involved in the formation of myelin, its homeostasis is regulated by several metabolic pathways and for its elimination is indispensable the participation of the Cholesterol 24 Hydroxylase (CYP46A1). This enzyme oxidize cholesterol to 24S-hydroxycholesterol for been exported through the blood brain barrier (BBB). In vitro studies have shown that high levels of cholesterol in the CNS may induced neurodegeneration, suggesting that a deficit of CYP46A1 could contribute to the development of neurodegenerative diseases. The aim of this study is to determine whether obesity modify cholesterol homeostasis into the CNS. Four groups of male Wistar rats were fed for 2, 4 and 6 months with different hypercaloric diets: 1. Normal diet, 2. High sucrose diet, 3. High fat diet, and 4. High sucrose+high fat diet. Our preliminary results showed that all hypercaloric diets induced obesity. The expression of CYP46A1 decreases in the hippocampus of the rats fed with the high sugar and high fat diet groups, but not in the high sucrose+high fat group. The low levels of CYP46A1 could contribute to the increase in cholesterol levels into CNS and thus to the development of neurodegeneration; feature that share neurodegenerative disorders such as Alzheimer's Disease, Parkinson's Disease, and others. These data are a preamble for the search of the molecular mechanism involved in association between obesity and neurodegenerative diseases.

Disclosures: **N. Gomez-Crisostomo:** None. **E. Martínez-Abundis:** None. **E. De la Cruz-Hernández:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.23/V19

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant 2R15NS060117-02

Title: Changes in hippocampal protein expression following diet supplementation of Kale, Arugula, or Dandelion in diet induced obese pre-diabetic C57BL/6 mice

Authors: *D. HICKS[1], A. A. OYETUNDE[1], D. FOSTER[2], B. TENG[1], L. R. BANNER[1]
[1]Biol., [2]California State Univ. Northridge, Northridge, CA

Abstract: Diabetes is a chronic disease that can lead to serious long-term complications. Type 2 diabetes has been associated with impaired cognitive performance and dementia. Correlations between increased body mass and an elevated risk for neurodegeneration and dementia have been documented. There is also evidence that links increased body mass to changes in hippocampal and spatial learning. These cognitive effects begin as early as childhood and adolescence in type 2 diabetes and include both structural and functional alterations in humans and animal models. Changes in diet are the most effective way to treat type 2 diabetes, however, 97% of dieters regain their lost weight within 3 years. Treatment with dietary supplements may prove a practical means by which to counter the cognitive impairment associated with diabetes. To examine the effect of these supplements, a high-fat diet (HFD) prediabetes mouse model was used. Obese and control mice given a dietary supplement of kale, arugula,dandelion for 22 weeks exhibited attenuated cognitive decline. The molecular mechanisms underlying this improvement will be analyzed by quantification of various marker proteins for neurogenesis, synaptic density, neuroinflammation, and aberrant brain insulin signaling in hippocampus. To this end, the expression of PSD95, syntaxin, GFAP, DCX, BDNF, IRS-1, and others will be compared by western blot. IL-1β, IL-6, and TNF-α will also be compared by ELISA. PSD95 and syntaxin are synaptic markers that localize to postsynaptic and presynaptic terminal respectively. GFAP is expressed in activated astrocytes in neuroinflammation. IL-1β, IL-6, and TNF-α are pro-inflammatory cytokines. While neuroinflammation is an important process in the healthy brain, chronic inflammation, as has been observed in diabetes and in obesity, can lead to tissue damage, neurodegeneration, and dementia. BDNF promotes the generation of new neurons and synapses. Decreased levels of BDNF have been implicated in various dementias and BDNF in plasma has been shown inversely correlated with blood glucose in diabetes. DCX is a marker of dividing neuronal precursor cells. IRS-1 is a substrate of brain insulin receptors whose abnormal

phosphorylation is associated with tau hyperphosphorylation seen in various diabetes-linked dementias including Alzheimer's. Furthermore, IRS-1 is upstream in a signaling pathway that phosphorylates GSK3β which can act as a tau kinase. Preliminary results show changes in the expression of PSD95, GFAP, and DCX in HFD-treated animals. Analysis of these, and other, molecular markers will shed light on the mechanism of diet-induced cognitive changes in type-2 diabetes.

Disclosures: D. Hicks: None. **A.A. Oyetunde:** None. **D. Foster:** None. **B. Teng:** None. **L.R. Banner:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.24/V20

Topic: C.01. Brain Wellness and Aging

Support: MJFF Grant 9524

NIH Grant P30MH062261

Title: Impaired synaptic mitochondrial spare respiratory capacity in aging and age-related neurodegenerative disease

Authors: *K. L. STAUCH, K. EMANUEL, B. LAMBERTY, B. MORSEY, H. FOX
Pharmacol. and Exptl. Neurosci., Univ. of Nebraska Med. Ctr., Omaha, NE

Abstract: Although studies have begun to elucidate the physical, biological, chemical, and psychological changes that occur during aging, the underlying molecular mechanisms involved in both healthy and pathological brain aging remain incompletely characterized. Mitochondrial dysfunction has been identified as a major player leading to age-associated alterations in the brain and has been implicated in the pathogenesis of age-related neurodegenerative diseases including Alzheimer's and Parkinson's. Additionally, as HIV+ individuals are living longer, the impact of aging on HIV-associated neurocognitive disorder and antiretroviral drug toxicity is an emerging challenge, as antiretrovirals and other drugs can affect mitochondria. Increasing evidence implicates mitochondrial malfunction as a trigger leading to synaptic failure and synaptic alterations in normal aging are linked to cognitive impairment. Therefore, we have examined the synaptic mitochondrial alterations that occur in animal models of aging and age-related neurodegenerative disease. Bioenergetic functional analysis revealed similar alterations in mitochondrial spare respiratory capacity between different disease states, which we propose underlies synaptic dysfunction. Further, we found that antiretroviral drug exposure perturbs this same functional parameter in synaptic mitochondria. This work highlights commonalities

between synaptic mitochondrial changes in different neurodegenerative disease states, providing a framework for future studies that will lead to improvements in the treatment of neurocognitive deficits in both age-related diseases and HIV.

Disclosures: K.L. Stauch: None. **K. Emanuel:** None. **B. Lamberty:** None. **B. Morsey:** None. **H. Fox:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.25/V21

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant

Title: Impact of chronic stress on sex-divergent CRF1 expression and behavioral deficits in APP/PS1 mouse model of Alzheimer's disease

Authors: *H. DONG[1], Y. YAN[2,1], G. RODRIGUEZ[1], R. GAO[1], J. G. CSERNANSKY[1]
[1]Dept Psychiatry and Behavioral Sci., Northwestern Univ. Feinberg Sch. of Med., Chicago, IL; [2]Physiol., Zunyi Med. Univ., Zunyi, China

Abstract: Alzheimer`s disease (AD), one of the most common neurodegenerative diseases, is characterized by progressive cognitive deficits and dementia. Accumulation of amyloid-beta protein plaques (Aβ) and hyperphosphorylated tau tangles are the pathological hallmarks of AD. Clinical studies have consistently demonstrated that women are disproportionately affected by AD with respect to both incidence and severity. Studies have shown that out of the 5.4 million American patients diagnosed with AD, 3.4 million, roughly 70%, are women. In addition, studies have found that the estimated lifetime risk for Alzheimer's specifically at age 65 was 17 percent for women and 9 percent for men. While clinicial studie cases have shown a sex gender difference in incedence, the biological mechanisms underlying this sexual divergence have not been well explored. In our previous work, we demonstrated that sex-specific biochemical differences in the central stress response, mediated by the corticotrophin releasing factor 1 receptor (CRF1) biases female mice towards pro-AD signaling on a phosphoproteomic level. In this study, we investigated whether chronic stress induced sex-divergent CRF/CRF1 downstream signaling pathways mediate AD pathology and memory function deficits in APP/PS1 mice. A total of 40 APP/PS1^{+} mice (20 males and 20 females) at 5 months of age were used for this study, with half exposed to chronic unpredictable mild stress (CUMS) and the other half without any stress for 21 days. Following the 21 days, a series of behavioral tests, including the tests for anxiety-like behavior and learning/memory function, were conducted. Brain tissues were subsequently collected for biochemical and neuropathological analysis of CRF1 and PKA

expression, as well as amyloid plaque measurement. We found that APP/PS1$^+$ female mice increased anxiety-like behavior and memory deficits as compared to APP/PS1$^+$ male mice after CUMS. Female mice also displayed more plaques in the cortex and hippocampus as compared to stressed males and non-stressed females. Moreover, in parallel with the sex biases behavioral and pathological changes, we found that stressed APP/PS1$^+$ female mice displayed higher expression of CRF1 and PKA in the hippocampus as compared to stressed APP/PS1$^+$ males and non-stressed females. Our results indicate during chronic stress, the CRF1 signaling pathway may display a sex-dependent response in regulation of neuronal function and neuropathogenesis of AD in an animal model.

Disclosures: H. Dong: None. **Y. Yan:** None. **G. Rodriguez:** None. **R. Gao:** None. **J.G. Csernansky:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.26/V22

Topic: C.01. Brain Wellness and Aging

Support: Max Planck Society

Yale University

NIH

Title: The role of Dnmt3a2 in regulating neuronal and age-associated cognitive functions

Authors: D. CUI[1], S. NEUBER[1], I. INGO VOIGT[1], Y. YAYOI OBATA[2], *X. XU[3,4]
[1]Epigenetic regulation of mammalian ageing, Max Planck Inst. For Biol. of Ageing, Cologne, Germany; [2]Dept. of Biosci., Tokyo Univ. of Agr., Tokyo, Japan; [3]Epigenetic regulation of mammalian aging, Max Planck Inst. For Biol. of Aging, Cheshire, CT; [4]Yale Univ. Sch. of Med., New Haven, CT

Abstract: Emerging evidence suggests that DNA methylation plays a crucial role in the pathogenesis of age-related neurological symptoms. Findings from our study and the literature indicate that DNA methyltransferase 3a2 (Dnmt3a2), an isoform of Dnmt3a, may be a key factor for maintaining normal hippocampal function and confers an epigenetic mechanism for age-related defects in cognitions. To test this hypothesis, Dnmt3a2 overexpression and knock-down neuronal cells, and the forebrain neuron-specific Dnmt3a2 transgenic (TG) mouse lines were generated and applied for the study. Our results show that the expression level of Dnmt3a2 in neuronal cells positively regulates the expression of synaptic plasticity genes and strengthens the cell resistance to stress insults by epigenetically modulating the transcription of neuronal genes

and cell apoptotic pathways. Moreover, we found that Dnmt3-like (Dnmt3L) protein can enhance the protective function of Dnmt3a2 in neuronal cells against the stress stimulus by increasing Dnmt3a2 nuclear localization. Meanwhile, the forebrain neuron-specific Dnmt3a2 and Dnmt3a2/Dnmt3L TG mouse lines were established and characterized, respectively. The cognitive behaviour measurement, cellular and molecular functional studies for both TG lines are ongoing. Taken together, our study will provide an epigenetic mechanism that DNA methylation contributes to neuronal and brain aging as well as the age-related deficit on learning, memory and cognition.

Disclosures: D. Cui: None. **S. Neuber:** None. **I. Ingo Voigt:** None. **Y. Yayoi Obata:** None. **X. Xu:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.27/V23

Topic: C.01. Brain Wellness and Aging

Support: National Science Foundation Graduate Research Fellowship Award

Ruth L. Kirschstein National Research Service Award (NRSA) Individual Predoctoral Fellowship (Parent F31)

Thomas Shortman Training Fund Scholarship Award

Axol Science Award

NIH

Robert Packard Center for ALS Research

ALS Association

Title: Nuclear pore complex composition in the mammalian cns: Regional and cell type specific differences

Authors: *J. C. GRIMA, S. VIDENSKY, A. N. COYNE, T. PHILIPS, J. G. DAIGLE, K. ZHANG, M. MATUNIS, J. D. ROTHSTEIN
The Solomon H. Snyder Dept. of Neurosci., The Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: Nuclear Pore Complexes (NPC) are large molecular structures that serve as the main gateways between the nucleus and cytoplasm. They not only directly control the exchange of

protein and RNA into and out of the nucleus but also serve transport-independent functions such as regulating genome organization and gene expression. The proper functioning and biogenesis of these large channels are critical for cell homeostasis and survival. NPCs consist of 30 different proteins called nucleoporins (NUPs) that are organized into five distinct anatomical regions of the NPC. Increasing evidence shows that each of these NUPs play very unique and specific roles. For instance, NUP50 serves a critical role in nuclear import, XPO1 plays an essential function in nuclear export, and GLE1 plays a significant part in mRNA export. Also, each NUP has been shown to traffic unique subsets of macromolecules thus adding another layer of diversity and specificity. One can begin to imagine that cells may be able to regulate certain cellular processes, given cell type-specific constraints and context-dependent needs, by altering the composition of NPCs and that this may provide an explanation for tissue-specific diseases that arise from mutations in various NUPs. For instance, mutations in NUP62 cause Infantile Bilateral Striatal Necrosis, the selective degeneration of the striatum in infants, and mutations in GLE1 cause Lethal Motoneuron Syndrome (LCCS1), a fetal motor neuron disease, as well as rare forms of Amyotrophic Lateral Sclerosis (ALS). It is possible that NUP62 is responsible for the trafficking of a transcription factor that is absolutely necessary for striatum viability. The same can be postulated for GLE1 and motor neurons. Although the overall structure of the NPC is conserved across different cell types, the aforementioned data suggests that protein composition of NPCs may vary among cell types and tissues. Lastly, very little is known about NPC structure and function in the mammalian CNS and whether brain cells use unique combinations of different NUPs to create NPCs bearing distinct properties and specialized functions. To this end, we have employed a novel method to isolate and purify NPCs from specific cell types and across different tissues of the CNS and characterize the composition of these NPCs using mass spectrometry and super resolution imaging analyses. We believe these ongoing experiments will provide critical insight into NPC neurobiology, which is relatively unknown. This will in turn provide a foundation for understanding CNS nucleocytoplasmic transport, which is a new and exciting focus for neurodegeneration pathobiology.

Disclosures: **J.C. Grima:** None. **S. Videnskу:** None. **A.N. Coyne:** None. **T. Philips:** None. **J.G. Daigle:** None. **K. Zhang:** None. **M. Matunis:** None. **J.D. Rothstein:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.28/V24

Topic: C.01. Brain Wellness and Aging

Support: RTC-2015-4094-1

EDU/346/2013

ULe Grant

Title: Age-dependent differences in TNFR1 response after global cerebral ischemia

Authors: *E. FONT BELMONTE[1], B. ANUNCIBAY SOTO[2], M. SANTOS GALDIANO[3], D. PEREZ-RODRIGUEZ[1], P. GONZALEZ-RODRIGUEZ[1], M. REGUEIRO-PURRIÑOS[4], A. FERNANDEZ-LOPEZ[1]
[1]Univ. de Leon, Inst. de Biomedicina, Leon, Spain; [2]Area de Biologia Celular, Univ. Leon. Inst. De Biomedicina, Leon, Spain; [3]Univ. de Leon. Inst. de Biomedicina, Leon, Spain; [4]Hosp. Veterinario de León. Univ. de León, León, Spain

Abstract: Age is one of the most important factor in the development of neurodegenerative or cerebrovascular diseases such as ischemic stroke, due to this, stroke is one of the most important cause of death in the elderly population. Unfortunately, the studies that reach neuroprotection in animal models of stroke, don't reach succeeded results in clinical assays. A possible explanation of the discrepancies between the research results and the clinical assays could be the different response to an ischemic insult that occurs as consequence to the age. However the majority of the studies are performed in young animals, which are not good models of the ischemic effects on the neural tissues in the elderly individuals. So how the age modifies the molecular response after a stroke is not clear. After a severe stress like an ischemic stroke, TNFR1 can elicit both a pro-survival NF-κB dependent signal or initiates regulated cell death that includes apoptosis or necroptosis. In the lasts years necroptosis has emerged as a different modality of regulated cell death, which can be used as a therapeutic target in several pathologies.Here we present an aging study comparing the response in young and old animals. We perform global cerebral ischemia model with 2 vessel occlusion combined by a partial exsanguination. In this report we analyzed the TNFR1 mediated response and the hallmarks of the necroptosis pathway 48h after the ischemia in young and aged animals in two areas which present different vulnerability to the ischemic damage (cerebral cortex and hippocampus). We measured by qPCR or Western blot changes in the receptor (TNFR1) and its associated ubiquitin-ligases (cIAP1), in the NF-κB transcriptional activity (IκB mRNA levels) and in the induction of regulated cell death process (MLKL, pMLKL, cFLIP). We conclude that aging enhance the necroptotic signal after ischemia, and can modify the inflammatory response exerting changes in the TNRF1 signaling and modifying the NF-κB transcriptional activity. This work was supported by the Spanish Ministerio de Economia y Competitividad (MINECO) cofinanced with FEDER funds ``una manera de hacer europa´´ RTC-2015-4094-1. P Gonzalez-Rodriguez, B Anuncibay-Soto and E Font-Belmonte are supported by Universidad de Leon. D. Perez-Rodriguez is granted by Junta de Castilla y Leon (EDU/346/2013).

Disclosures: E. Font Belmonte: None. **B. Anuncibay Soto:** None. **M. Santos Galdiano:** None. **D. Perez-Rodriguez:** None. **P. Gonzalez-Rodriguez:** None. **M. Regueiro-Purriños:** None. **A. Fernandez-Lopez:** None.

Poster

385. Brain Wellness and Aging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 385.29/V25

Topic: I.02. Systems Biology and Bioinformatics

Support: Perelman School of Medicine at the University of Pennsylvania

Fudan University

Title: Stroke 1-2-0 fills the gap for prehospital delay for stroke victims in China

Authors: *J. ZHAO
Fudan University,Minhang Hosp., shanghai, China

Abstract: Background
Although intravenous thrombolytic therapy is available in China, studies indicated that only 1.6-4.0 % of ischemic stroke patients received such thrombolytic therapy due to significant prehospital delay with the median time of the delay has 15 hours.[1] To reduce prehospital delay in China, we engineered stroke 120, a stroke educational program suitable for China for a rapid stroke recognition and quick response. This study examined whether this novel tool is well accepted in China via online survey and onsite survey. This investigation protocol was approved by the Institutional Review Board of the Minhang Hospital affiliated to Fudan University. There were total of 927 people from 30 different regions (province or cities) participated online survey. 45% are males and 55% are females. 60% of the responders had family member, relative or friends who had stroke in the past. While majority of the people (85%) recognize that it is critical to identify stroke and transport patients to hospitals, knowledge for the timing of the therapy and which hospital should the patient sent to are very deficient. 49% thought that stroke 1-2-0 is best suitable for China for stroke awareness and rapid response, 21% thought that FAST (“Face”, “Arm”, “Speech”, “Time”) should be used. 61% of the survey responders thought that a consent is needed for thrombolitic therapy. The onsite survey obtained similar results in 584 participants, majority of the participants thought that Stroke 120 is the best program to be used for stroke awareness education, 18% of them thought FAST is the best program. Based on this survey results, it clearly indicated that stroke 1-2-0 is suitable for and is well accepted in China.

References

1

Disclosures: J. Zhao: None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.01/V26

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH U54 AG054345-01

Title: Model-ad: Genetic models of late-onset Alzheimer’s disease

Authors: *A. OBLAK[1], G. CARTER[4], G. R. HOWELL[5], B. LOGSDON[6], L. MANGRAVITE[6], K. NHO[2], L. OMBERG[6], K. D. ONOS[5], V. PHILIP[4], C. PREUSS[4], S. J. SUKOFF RIZZO[4], M. SASNER[4], L. SHEN[2], A. J. SAYKIN[3], P. TERRITO[2], A. UYAR[4], H. WILLIAMS[4], B. T. LAMB[7]
[1]STARK Neurosci. Res. Inst., [2]Radiology & Imaging Sci., [3]Radiology and Imaging Sci., Indiana Univ. Sch. of Med., Indianapolis, IN; [4]The Jackson Lab., Bar Harbor, ME; [5]Jackson Lab., Bar Harbor, ME; [6]Sage Bionetworks, Seattle, WA; [7]Stark Neurosciences Res. Inst., Indianapolis, IN

Abstract: Alzheimer’s disease (AD) is the most common form of dementia, with no effective preventative strategies or treatments. One of the major obstacles of developing novel therapies for AD is the lack of predictive animal models to be used in preclinical trials of therapeutics. One reason for this may be that existing models are based on familial mutations, while the vast majority of the clinical population has non-familial late-onset AD (LOAD). The Model Organism Development and Evaluation for Late-onset AD (MODEL-AD) Center has been established as a consortium consisting of Indiana University, The Jackson Laboratory, and SAGE Bionetworks. The goals of the Center are: to identify novel genetic variants, genes and biomarkers from AD patient data; to generate and validate new animal models based on these LOAD variants; and to utilize these novel models in a preclinical testing paradigm. The Bioinformatics and Data Management Core (BDMC) is prioritizing novel sequence variants, creating analytical pipelines for human-mouse phenotype comparisons, and analyzing phenotypic data. Biomarkers and disease model endophenotypes will be compared to patient data wherever possible. All data will be made available through the SAGE-Synapse portal. The Disease Modeling Project (DMP) is creating new mouse models based on variants identified by the BDMC. *APOE4* and *Trem2* alleles are the strongest genetic risk factors for LOAD; as such we have created a novel model expressing both human *APOE4* and the R47H allele of *Trem2*. This is being phenotypically characterized using functional assays, neurodegeneration, amyloid and tau pathology, transcriptional and metabolic profiling, and *in vivo* imaging. Existing familial models (5XFAD, APP/PS1, and hTau) are being evaluated for comparative purposes. The *APOE4/Trem2* model will serve as the standard background as we introduce additional LOAD genetic variants, including *ABCA7*, *IL1RAP* and *CR1*. Humanized *APOE3* and *APOE2* models,

to be used as controls, are also being generated. The Preclinical Testing Core (PTC) has established a tertiary screening pipeline with predetermined go/no go criteria to evaluate novel compounds in new models that have been shown to have an AD-like phenotype. These criteria include exposure levels in target tissues, target engagement and disease modifying effect, and *in vivo* functional activity and therapeutic index. The pipeline is currently being validated using levetiracetam in the 5XFAD model. All models, protocols, and data sets are to be made widely available. We seek input and collaborations from the research and pharma/biotech communities. For more information see www.model-ad.org.

Disclosures: **A. Oblak:** None. **G. Carter:** None. **G.R. Howell:** None. **B. Logsdon:** None. **L. Mangravite:** None. **K. Nho:** None. **L. Omberg:** None. **K.D. Onos:** None. **V. Philip:** None. **C. Preuss:** None. **S.J. Sukoff Rizzo:** None. **M. Sasner:** None. **L. Shen:** None. **A.J. Saykin:** None. **P. Territo:** None. **A. Uyar:** None. **H. Williams:** None. **B.T. Lamb:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.02/W1

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: U54 AG054345-01

Title: Preclinical drug screening in new generation Alzheimer’s disease mouse models: The MODEL-AD Consortium Strategy

Authors: *P. R. TERRITO[1], S. J. SUKOFF RIZZO[3], K. D. ONOS[4], J. A. MEYER[1], J. PETERS[1], S. PERSOHN[1], B. MCCARTHY[1], A. RILEY[1], S. QUINNEY[2], D. JONES[2], M. SASNER[3], G. R. HOWELL[3], H. WILLIAMS[3], A. OBLAK[1], B. T. LAMB[5]
[1]Radiology and Imaging Sci., [2]Indiana Univ. Sch. of Med., Indianapolis, IN; [3]The Jackson Lab., Bar Harbor, ME; [4]Jackson Lab., Bar Harbor, ME; [5]Stark Neurosciences Res. Inst., Indianapolis, IN

Abstract: Historically, preclinical screening of test compounds for Alzheimer’s Disease (AD) employed behavioral endpoints in rodent models as the primary screen. Often the rodent models used did not necessarily have construct validity for AD, and experiments often evaluated the ability of a test compound to reverse an acute pharmacological deficit (e.g. scopolamine induced memory deficit) in wild-type or normal animals. Other screens evaluated the ability of the test compound to normalize a behavioral phenotype, and these studies rarely used biomarkers or other clinically translational endpoints. Young or naïve wild-type animals were often used in place of aging animals, and critically pharmacokinetic (PK) and pharmacodynamic data in the AD model at the biologically and pathologically relevant ages had not been evaluated. The

Model Organism Development and Evaluation for Late-onset AD (MODEL-AD) program has been established with the goals of: 1) identifying novel genetic variants, genes and biomarkers from LOAD patient data; 2) generating and validating mouse models with construct and face validity for LOAD; and 3) developing a preclinical testing strategy to evaluate potential therapeutic agents for the treatment of AD in these new models. The Preclinical Testing Core (PTC) has established a streamlined preclinical strategy with go/no-go decision points that allow critical and unbiased assessments of potential therapeutic agents while matching each test compounds' specific mechanism of action to the animal model best suited to interrogate its symptom and/or disease modifying activity. The PTC screening strategy includes an initial primary screen to determine appreciable mutli-dose PK and target tissue activity in the disease model at the pathologically relevant age followed by predictive PK/PD modeling. A secondary screen evaluating target engagement and disease modifying activity of the test compound utilizing non-invasive PET/MRI as a pharmacodynamic readout of cerebral changes in metabolism (18F-FDG), cerebral blood flow (64Cu-PTSM), beta amyloid deposition (18F-AV45), and tau deposition (18F-AV1451). Provided the compound meets the a priori criteria for success in the primary and secondary screens, the tertiary screen will evaluate symptom modifying effects of the test compound to normalize a disease-related phenotype in cognition tests, relative to the age- and sex-matched littermate WT controls. All raw data, standard operating procedures, methods and protocols will be made publicly accessible via the external facing SAGE portal to the AD research community at large.

Disclosures: P.R. Territo: None. **S.J. Sukoff Rizzo:** None. **K.D. Onos:** None. **J.A. Meyer:** None. **J. Peters:** None. **S. Persohn:** None. **B. McCarthy:** None. **A. Riley:** None. **S. Quinney:** None. **D. Jones:** None. **M. Sasner:** None. **G.R. Howell:** None. **H. Williams:** None. **A. Oblak:** None. **B.T. Lamb:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.03/W2

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant AG054345

Title: Novel candidate loci for Alzheimer's disease from whole-genome and whole-exome sequencing

Authors: *G. CARTER[1], X. WANG[2], C. PREUSS[1], V. PHILIP[1], G. ANANDA[3], C. J. ACKLIN[1], K. KARUTURI[3], M. SASNER[1], G. R. HOWELL[1]
[1]The Jackson Lab., Bar Harbor, ME; [2]Eisai Andover Innovative Medicines (AiM) Inst., Andover, MA; [3]The Jackson Lab. for Genomic Med., Farmington, CT

Abstract: Recent initiatives such as the Alzheimer's Disease Sequencing Project (ADSP) have created sequencing-based study cohorts for late-onset Alzheimer's disease. While this resource provides nucleotide-level resolution of genetic variants, it suffers from relatively small sample size and population substructure that limits association power. We addressed this limitation by devising a method of genome-wide association that implements a generalized linear mixed model in a Bayesian context. Our approach, Bayes-GLMM, has four main features: (1) support of categorical, binary and quantitative variables; (2) cohesive integration of previous GWAS results for related traits; (3) correction for sample relatedness by mixed modeling; and (4) model estimation by both Markov chain Monte Carlo (MCMC) sampling and maximal likelihood estimation. We applied Bayes-GLMM to the ADSP whole-genome sequencing cohort, which includes categorical disease diagnoses of 570 individuals drawn from 111 families. We also analyzed ADSP whole-exome data from 9134 individuals to identify rare and common coding variants. From the whole-genome sequencing cohort, we identified four variants in three loci significantly associated with Alzheimer's disease. The loci were not identified using traditional methods. The four variants (rs10490263, rs74944275, rs149372995, rs140233081) are located in intergenic regions with the closest genes not previously associated with AD. The two linked variants lie between the genes *PRKAR1B* and *PDGFA*. These proteins are localized to the glial-vascular unit, further implicating vascular function in modifying susceptibility to AD. Analysis of whole-exome sequences validated association of many known loci (e.g. *TREM2, ABCA7*) and identified potentially novel variants, including both rare and common alleles. This work provides new candidates loci for Alzheimer's disease, obtained from the first implementation of a flexible, generalized mixed model approach in a Bayesian framework for sequence-based association studies.

Disclosures: G. Carter: None. **X. Wang:** A. Employment/Salary (full or part-time):; Eisai. **C. Preuss:** None. **V. Philip:** None. **G. Ananda:** None. **C.J. Acklin:** None. **K. Karuturi:** None. **M. Sasner:** None. **G.R. Howell:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.04/W3

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant U54 AG054345-01

Title: Model-ad: Bioinformatics and data management core

Authors: *B. T. LAMB[1], M. SASNER[2], A. J. SAYKIN[3], L. MANGRAVITE[4], G. CARTER[2]
[1]Stark Neurosciences Res. Inst., Indianapolis, IN; [2]The Jackson Lab., Bar Harbor, ME;

[3]Radiology and Imaging Sci., Indiana Univ. Sch. of Med., Indianapolis, IN; [4]Sage Bionetworks, Seattle, OR

Abstract: Creation of the next generation of animal models in the Model Organism Development and Evaluation for Late-onset Alzheimer's Disease (MODEL-AD) consortium will require extensive data analysis, curation, and distribution. To this end, a central Bioinformatics and Data Management Core (BDMC) has been created to integrate information from large-scale data resources including the Alzheimer's Disease Sequencing Project, Alzheimer's Disease Neuroimaging Initiative, the Accelerating Medicines Partnership for Alzheimer's Disease (AMP-AD), Molecular Mechanisms of Vascular Etiology of Alzheimer's Disease, and others. We are creating bioinformatic pipelines to: (1) identify key genetic variants that confer risk of AD from genome-wide association studies; (2) translate candidate variants into mouse models; (3) align human disease and animal model phenotypes to specify the optimal research use of each animal and characterize the effects of multiple genetic variants; and (4) broadly disseminate all data and preclinical research protocols for community use. Here we present our early results and future plans to meet these needs. To date, multiple variants have been identified through the integration of results from multiple genetics studies, at both novel and known genetic loci. Genomic, epigenetic, and functional data have been used to predict the consequences of candidate variants, thereby prioritizing specific genetic alterations for animal modeling via genetic engineering. Molecular phenotypes, including transcriptomics data from the brains of mouse models, have been systematically compared to similar data from clinical cohorts to identify pathways altered in both human and mouse. Finally, we are broadly sharing data via the AMP-AD Knowledge Portal hosted by Sage Bionetworks. Future work will expand all of these findings and resources, and additionally integrate data and protocols from preclinical studies that use the animal models. In sum, the BDMC is creating a resource to integrate a broad knowledgebase to serve the creation and study of multiple animal models to effectively model late-onset Alzheimer's disease *in vivo*.

Disclosures: **B.T. Lamb:** F. Consulting Fees (e.g., advisory boards); Eli Lilly, Amgen, Abbvie. **M. Sasner:** None. **A.J. Saykin:** None. **L. Mangravite:** None. **G. Carter:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.05/W4

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: U54 AG054345-01

Title: MODEL-AD: The disease modeling project

Authors: *M. SASNER[1], A. OBLAK[2], H. WILLIAMS[1], G. HOWELL[1], B. T. LAMB[2]
[1]The Jackson Lab., Bar Harbor, ME; [2]Stark Neurosciences Res. Inst., Indianapolis, IN

Abstract: The Alzheimer's Disease (AD) patient population consists almost entirely (~98%) of the late-onset form of AD (LOAD); however, most mouse models used to study AD are based on familial AD mutations in *APP*, *PSEN1* or *PSEN2*. The Model Organism Development and Evaluation for Late-onset AD (MODEL-AD) Center was created to develop, characterize, and distribute more precise preclinical models for AD.
The Disease Modeling Project (DMP) of MODEL-AD will use CRISPR genome editing to generate at least 40 novel mouse models that carry various combinations of human risk alleles for LOAD that have been identified by the Bioinformatics and Data Management Core (BDMC). In the early years of the Center, we will prioritize understanding known GWAS variants (*ABCA7* and *CR1*) as well as novel variants identified by BDMC analyses of the AD Neuroimaging Initiative dataset (*IL1RAP*).
We will utilize a two-phase screening strategy. At least 24 models will undergo high capacity screening at 12 months of age, which includes functional assays and AD-related pathology. The most promising models will advance to the deep phenotyping phase, which will occur independently at Indiana University and The Jackson Laboratory to ensure reproducibility of data. In the deep phenotype screening, cohorts of mice will be evaluated at three time points (2, 6, and 12 months). We will prioritize clinically relevant endpoints including *in vivo* imaging, blood and tissue biomarkers, and genomics, and compare these to more traditionally used endpoints such as battery of functional assays. All data will be provided to the BDMC to determine the predictive potential for each mouse model, which will then be prioritized and selected for the preclinical testing core (PTC). To test our pipeline, we will deep phenotype models currently used for studying familial AD (APP/PS1, 5XFAD, hTau). In addition, we will also deep phenotype a new model of LOAD that we have created. This model carries the two greatest genetic risk factors for LOAD, *APOE4* and the R47H variant of *TREM2*. Our strategy closely integrates human and mouse data, with the aim that these new AD models will show a high degree of clinical translatability for preclinical testing of new therapeutic targets.
All models created will be made available at the earliest opportunity through the JAX AD Mouse Mutant Resource. All protocols and data will be made available via the SAGE Synapse portal (www.synapse.org); we anticipate that we will be able to provide not only improved models, but also validation protocols and data so that the research community can efficiently adopt these new models. For more information, see *model-ad.org.*

Disclosures: M. Sasner: None. **A. Oblak:** None. **H. Williams:** None. **G. Howell:** None. **B.T. Lamb:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.06/W5

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: VASDHS

Title: Comparison of gene expression profile in Alzheimer's disease using a novel stratifying algorithm

Authors: *S. CANCHI[1], R. SASIK[2], P. D. JAGER[3,4], D. A. BENNETT[4], R. A. RISSMAN[5]
[1]Univ. of California San Diego, LA Jolla, CA; [2]Univ. of California San Diego, La Jolla, CA; [3]Ctr. for Translational & Computat. Neuroimmunology, Dept. of Neurol., Columbia Univ., New York, NY; [4]Rush Alzheimer's Dis. Ctr., Rush Univ. Med. Ctr., Chicago, IL; [5]Neurosci, UCSD Sch. Med., La Jolla, CA

Abstract: The discovery of mutations in *Amyloid precursor protein (APP), Presenilin 1 (PSEN1),* and *Presenilin 2 (PSEN2)* as a cause of autosomal dominant Alzhiemer's disease (AD) and the identification of the ε4 allele of *Apolipoprotein E* (*APOE*) as strong genetic risk factor have been instrumental in understanding the pathological processes underlying AD; the findings from GWAS re-emphasize the multifactorial nature of AD dementia. Despite these advances, translation of genetic findings into functional mechanisms that are biologically important in disease pathogenesis and treatment design remains a challenge. Using a novel stratifying algorithm 640 subjects from the Religious Orders Study (ROS) and the Rush Memory and Aging Project (MAP) cohorts were segregated into six relevant diagnosis groups. Expression data (fpkm) were first normalized using a multi-loess method and batch effect was removed using ComBat implemented in the *R* package *sva*. We were interested in genes that are differentially expressed between the diagnostic groups controlling for the covariates sex, age, and ApoE status. We modeled the (log-)expression of a gene as a linear combination of sex, age, ApoE status, and diagnosis, using the *R* package *limma*. Formally, expression~0+sex+ApoE+age+diagnosis. Tests for significant differences between pairs of diagnoses were obtained by implementing a Bayesian strategy, *eBayes*. For each pair of diagnoses (the 15 comparisons) we sorted the genes by their posterior error probability (PEP). In each comparison, the difference between diagnosis effects equals the natural log of expression ratio between the two diagnoses, adjusted for other co-variates. Compared to the control subjects, 1410 genes were differentially expressed in the high probability AD group, 692 genes in the intermediate probability AD group and 43 genes in the Non-AD dementia group as defined by the modified NIA classification. Functional enrichment for the number of significant genes (PEP < 0.1) in all comparisons conducted using ToppGene revealed distinct pathways and gene families implicated in AD when compared to controls and

Non-AD dementia. The results are further discussed in the light of conventional diagnosis of AD dementia which help identify the differences in the transcriptome signature by diagnostic group.

Disclosures: S. Canchi: None. **R. Sasik:** None. **P.D. Jager:** None. **D.A. Bennett:** None. **R.A. Rissman:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.07/W6

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01 NS42818

Title: Conditional deletion of APP, APLP1, and APLP2 in excitatory neurons of the postnatal forebrain does not cause neurodegeneration

Authors: *J. KANG, A. HO, H. WATANABE, J. SHEN
Harvard Med. School/Bwh, Boston, MA

Abstract: The amyloid precursor protein (APP) has been implicated in both familial and sporadic Alzheimer's disease (AD). Despite its importance in AD pathogenesis, the study of its normal physiological role has been complicated by functional redundancy of its family members, the amyloid precursor protein like 1 and 2 (APLP1 and APLP2), and by the perinatal lethality of germline inactivation of *APP/APLP1/APLP2*. To determine the normal physiological role of the APP family, we generated postnatal forebrain-restricted *APP/APLP1/APLP2* conditional knockout (cKO) mice, in which the *APP*, *APLP1* and *APLP2* genes are deleted by Cre recombinase expressed under the control of the *αCaMKII* promoter in excitatory neurons. Despite the abundance of APP, APLP1 and APLP2 in excitatory neurons of adult cerebral cortex, their inactivation does not cause age-dependent neurodegeneration, as evidenced by the normal cortical volume and neuron number up to 21 months of age in *APP/APLP1/APLP2* cKO mice relative to littermate controls. In addition, *APP/APLP1/APLP2* cKO mice at 3 months of age exhibit mild learning and memory deficits in the Morris water maze, as shown by significant increases of latency during training and reduced quadrant occupancy in post-training probe trial, but they perform normally in the open field and rotarod tests. Furthermore, RNA-seq analysis of the hippocampal transcriptome of *APP/APLP1/APLP2* cKO mice at 3 months revealed significant alterations in mRNA levels of 305 genes with increases of 104 genes and decreases of 201 genes, consistent with a potential function of APP family in transcriptional regulation. Some of these alterations have been confirmed by quantitative RT-PCR and protein analysis; for example, calbindin1 mRNA and immunoreactivity are decreased in hippocampus of *APP/APLP1/APLP2* cKO mice. These findings provide insights into normal physiological

function of APP family and demonstrate that APP family is not required for cortical neuronal survival during aging.

Disclosures: J. Kang: None. **A. Ho:** None. **H. Watanabe:** None. **J. Shen:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.08/W7

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R03

Title: Genome-wide association analysis of amyloid-β and tau tangle deposition identifies enrichment of neurogenesis-related pathways

Authors: *E. HORGUSLUOGLU-MOLOCH[1], K. NHO[2], S. RISACHER[2], A. J. SAYKIN[3]
[1]Med. and Mol. Genet., [2]Radiology and Imaging Sci., Indiana University, Sch. of Med., Indianapolis, IN; [3]Radiology and Imaging Sci., Indiana Univ. Sch. of Med., Indianapolis, IN

Abstract: Introduction: Alzheimer's disease (AD) is characterized by the presence of senile (amyloid) plaques and neurofibrillary tangles. Tau plays a crucial role for neurogenesis and synaptic maturation of newborn hippocampal granule neurons and the absence of tau can result in retarded neurogenesis and neuronal differentiation (Pallas-Bazarra et al, 2016). While amyloid-beta (Aβ) accumulation has a negative effect on adult neurogenesis, Aβ precursor protein (APP) plays an important role in neuronal survival and maturation (Mu et al, 2011). To investigate whether tau tangles and Aβ deposition are significantly associated with neurogenesis-related pathways, we performed gene-set enrichment analysis on a genome-wide association study (GWAS) from the Alzheimer's Disease Neuroimaging Initiative (ADNI). **Methods:** Using non-Hispanic Caucasian participants from the ADNI cohort, GWAS was conducted using CSF levels ((N=975) $A\beta_{1\text{-}42}$, (N=1,154) t-tau, and p-tau_{181p}) and global cortical tau and Aβ deposition from [^{18}F] AV-1451 Flortaucipir tau (N=99) and [18F] florbetapir (N=800) PET scans as phenotypes controlling for age, sex and diagnosis. GSA-SNP was used to identify pathways enriched against Aβ and tau accumulation. **Results:** We identified 47, 54, and 43 pathways with enrichment for CSF t-tau levels, CSF p-tau_{181p} levels, and tau PET accumulation, respectively (FDR-corrected *p*-value < 0.05). 45 pathways were common with CSF t-tau and p-tau_{181p} levels, and related to neurogenesis, neuron differentiation, axonogenesis, neuron/neurite development, cellular differentiation, generation of neurons, and kinase activities. Of 45 enriched pathways, 12 pathways also were enriched for tau PET accumulation. . For 1,272 genes linked to the 45 common pathways enriched with CSF t-tau and p-tau181p levels, a gene-based association analysis identified APOE, PVRL2, and APOC4 as significantly associated with CSF t-tau and p-

tau181p levels after multiple comparison adjustment. In addition, Gene-set enrichment analysis of CSF $A\beta_{1-42}$ level and PET Aβ deposition identified 36 common significantly enriched biological pathways, which were related to neurogenesis processes including generation of new neurons, neuronal development, and neuronal migration and differentiation. **Conclusion:** Neurogenesis-related pathways which drive neural stem cell proliferation, maintenance in the adult neurogenic niche, and differentiation into mature neurons, were significantly enriched for tau and amyloid-β biomarker endophenotypes. The identification of enriched pathways contributes to the understanding of biological processes related to AD pathophysiology.

Disclosures: E. Horgusluoglu-Moloch: None. **K. Nho:** None. **S. Risacher:** None. **A.J. Saykin:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.09/W8

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: U54

Title: Characterizing neuronal pathology in the 5XFAD model of alzheimer's disease

Authors: *D. TUMBLESON-BRINK[1], S. PUNTAMBEKAR[2], S. M. BEMILLER[3], A. OBLAK[4], G. E. LANDRETH[5], B. T. LAMB[6]
[1]Med. and Mol. Genet., Indiana University-Purdue University,Indianapolis, Plainfield, IN; [2]Indiana University-Purdue Univ., Indianapolis, IN; [3]Neurosciences, IU Sch. of Med. Stark Neurosciences Res., Indianapolis, IN; [4]STARK Neurosci. Res. Inst., [5]Stark Neurosci. Res. Institute, NB214C, Indiana Univ. Sch. of Med., Indianapolis, IN; [6]Stark Neurosciences Res. Inst., Indianapolis, IN

Abstract: The Alzheimer’s Disease (AD) patient population consists almost entirely of the late-onset form of AD; However, there is a paucity of animal models that reflect the entire, pathological spectrum of Alzheimer’s Disease (AD). The Model Organism Development and Evaluation for Late-onset AD (MODEL-AD) Center was created to develop, characterize and distribute more precise preclinical models for AD. To this end, we are phenotyping the age related progression of neuronal pathology in one of the most commonly utilized mouse models of AD, the 5XFAD mouse model. These mice carry a total of five mutations in two of the genes that result in familial AD, *amyloid precursor protein (APP)* and *presenilin 1 (PS1)*, which leads to an overproduction of Aβ. This not only incrementally increases Aβ plaque deposition, but also results in neuron loss and cognitive deficits. Since our histological analysis of amyloid deposition highlighted the cortex, hippocampus, thalamus and the medulla as the primary regions

where pathology develops at 4 months of age, we chose to characterize neuron pathology in these 4 regions of interest. Using AT8 and AT180 to label hyperphosphorylated Tau epitopes and LAMP1 to identify neuronal dystrophy, distinct patterns of neuronal pathologies, namely inclusions of hyperphoshorylated tau in neuronal soma and/or axons and dystrophic neurites are observed. Histological labeling of AT8+ neurons shows the presence of AT8+ neuronal inclusions limited to the superficial layers of the prefrontal and frontal cortex and in the soma of neurons of the CA3, with no axonal labeling. However, predominant AT8 pathology is observed as dystrophic neurites in the deeper cortical layers, hippocampus, subiculum, thalamus and the medulla. Conversely, the primary pathology seen with AT180 is the presence of intraneuronal inclusions in all regions of interest, with very rare AT180+ dystrophic neurites. Interestingly, AT180+ axonal labeling is prevalent and restricted to the deep cortical layers. Histology with LAMP1 shows a complete absence of LAMP1+ dystrophic neurites in the superficial layers of the cortex with extensive LAMP1 labeling in the deep cortical layers as well as in the other regions of interest. This data is the first to describe a region-specific distribution of distinct neuronal pathologies in the 5XFAD model.

Disclosures: D. Tumbleson-Brink: None. **S. Puntambekar:** None. **S.M. Bemiller:** None. **A. Oblak:** None. **G.E. Landreth:** None. **B.T. Lamb:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.10/W9

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Gene expression and methylation analysis of ABCA7 in patients with Alzheimer’s disease

Authors: *K. YAMAZAKI
Ehime Univ. Hosp., Ehime, Japan

Abstract: *Background/Aim*: The aim of this study was to examine the blood gene expression and methylation of ATP-binding cassette sub-family A member 7 gene (ABCA7) as a biological marker of AD. ***Methods*:** AD subjects (n = 50; 11 males, 77.7 ± 6.05 years old) and age- and sex-matched healthy controls (n = 50) were recruited. A single nucleotide polymorphism in *ABCA7* (rs3764650), methylation rates of CpG sites in the *ABCA7* promoter region, and *ABCA7* mRNA expression levels in peripheral blood were examined. ***Results*:** The distribution of the rs3764650 polymorphism in AD subjects was not different from that of controls. Although the methylation rates of AD subjects were not significantly different from those of controls, the *ABCA7* mRNA expression level in AD subjects was significantly higher than that in controls. Additionally, the *ABCA7* mRNA expression level in AD subjects was significantly correlated with Mini Mental State Examination recall, the Alzheimer’s Disease Assessment Scale total score, and the Clinical

Dementia Rating score. We also found a significant correlation between the *ABCA7* mRNA expression level and duration of illness. ***Conclusion*:** The *ABCA7* mRNA expression level in peripheral blood may be a marker for early stages of AD and disease progression regardless of rs3764650 and the methylation rate of its promoter.

Disclosures: K. Yamazaki: None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.11/W10

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: ANZ Trustees Judith Jane Mason & Harold Stannett Williams Memorial Foundation

Alzheimer's Australia Dementia Research Foundation

JO and JR Wicking Trust

The Yulgilbar Foundation

Title: Charting epigenetic reprogramming of gene regulatory elements in AD mice

Authors: *A. J. PHIPPS[1], M. D. ROBINSON[4], J. C. VICKERS[2], T. R. MERCER[5], A. WOODHOUSE[2], P. C. TABERLAY[3]
[2]Wicking Dementia Res. and Educ. Ctr., [3]Sch. of Med., [1]Univ. of Tasmania, Hobart, Australia; [4]Inst. of Mol. Life Sci., Univ. of Zurich, Zurich, Switzerland; [5]Transcriptomics Res. Laboratory, Genomics and Epigenetics Div., Garvan Inst. of Med. Res., Sydney, Australia

Abstract: Introduction: Alzheimer’s disease (AD) is a terminal progressive neurodegenerative disorder, yet the underlying cause of sporadic AD cases remains unknown. Epigenetics allows for dynamic regulation of chromatin structure, and complex interaction between the DNA and our environment. Epigenetic alterations such as DNA methylation and post-translational histone modifications may contribute to the pathological pathways implicated in the onset and progression of sporadic AD. **Methods**: APP/PS1 AD mice closely recapitulate the pathology present in human early-AD cases, including beta-amyloid plaque deposition and plaque-associated synapse loss. APP/PS1 mice enable us to examine the earliest pre-pathology epigenetic changes that occur in AD, as well as a time course of disease progression. Neuronal nuclei from the forebrain of APP/PS1 mice and age-matched wild-type control mice (n=5 per genotype at 3,6,12 months of age) were isolated, then purified with FACS and subject to chromatin immunoprecipitation and next-generation sequencing (ChIP-seq) with antibodies detecting H3K4me3, H3K27me3, and H3K27Ac. **Results**: Enhancer (H3K27Ac) and promoter

(H3K4me3, H3K27me3) marks are lost from regulatory regions in APP/PS1 mice compared to age-matched controls ($p<0.05$). Specifically, both enhancer and promoter marks were lost from key risk factor genes for sporadic AD (*PICALM, BIN1*; $p<0.05$) or at known differentially expressed genes in sporadic human cases and transgenic AD mice (*PICALM, TBXA2R, F2RL2, SORBS3*; $p<0.05$). **Conclusions:** These data show that epigenetic reprogramming occurs in a mouse model of early AD, affecting both enhancers and promoters. Epigenetic dysregulation may be a key aspect of AD onset and pathology.

Disclosures: A.J. Phipps: None. **M.D. Robinson:** None. **J.C. Vickers:** None. **T.R. Mercer:** None. **A. Woodhouse:** None. **P.C. Taberlay:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.12/W11

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: AG040060

EB020403

Title: Towards multivariate genetic models predicting phenotypes of Alzheimer's disease risk

Authors: *B. C. RIEDEL[1], N. JAHANSHAD[2], D. A. BENNETT[3], P. M. THOMPSON[4]
[1]Neurosci., USC, Los Angeles, CA; [2]Neurol., IGC-INI @ USC, Marina Del Rey, CA; [3]Rush Univ., Chicago, IL; [4]Stevens Inst. for Neuroimaging & Informatics, Univ. of Southern California (USC), Marina del Rey, CA

Abstract: Alzheimer's disease (AD) has a variable phenotype and strong genetic component, indicative of multifactorial etiologies and trajectories. Apolipoprotein E (APOE) is the most established susceptibility gene for AD. Twin studies estimate heritability at up to 80%, yet APOE accounts for under 1/3 of this, and is not necessary to cause AD. Genome-wide association studies (GWAS) identify over 30 AD susceptibility genes, with most clustering in biological pathways such as lipid or glucose metabolism, yet many factors remain unknown. AD risk loci are likely difficult to replicate due to weak effects, heterogeneity, or interactions with other variants. We hypothesized the missing component in understanding genetic contributions to AD-risk require moving beyond univariate associations towards multivariate genetic models. Using the GWAS Catalog to identify single nucleotide polymorphisms (SNPs) with replicated association with AD, and the ENDEAVOR prioritization tool trained on the top 10 replicated AD-risk genes, we identified 37 AD risk genes showing evidence of systems interactions in mediating risk. After identifying multiple AD datasets with GWAS, data was filtered by

ethnicity to focus on individuals of western European descent. Data was combined across sites and filtered by allele frequency within the genes and surrounding upstream/downstream regions, Hardy-Weinberg equilibrium, and missingness. Top SNPs were determined using the fixation index and D Jost difference scores between controls and AD populations. Using the 12 SNPs across 7 genes identified, the ideal number of clusters with the Bayesian Information Criterion in the AD cohorts was found to be four. Clusters represent different genetic phenotypes associated with AD. APOE was associated with AD in 3 of the clusters. However, one cluster showed the least genetically driven profile and was not associated with APOE4. Ongoing work is exploring interactions using peripheral and brain gene expression arrays, RNA-seq, and structural brain imaging to create systems level insights into genetic phenotypes of AD risk.

Demographic Characteristics of AD and Healthy Controls					
		Total	Age (Mean ±SD)	Education	APOE4+
Controls	*N = 2638*		79.3 (8.6)	15.6 (3.3)	31%
	Males	1232	81 (8.4)	16 (3.2)	29%
	Females	1260	77.3 (8.4)	15.3 (3.3)	33%
AD	*N = 2371*		77.6 (9.8)	14.5 (3.7)	63%
	Males	982	80.1 (10.6)	15 (3.5)	58%
	Females	1117	75.7 (8.5)	14 (3.7)	68%

Table 1. Combined demographics of the four cohorts included in the study. These cohorts are the Religious Orders Study, the Rush Memory and Aging Study, the Harvard Brain Bank, the Alzheimer's Disease Neuroimaging Initiative, and the Late onset Alzheimer's Disease Family Study.

Genes Included in Multivariate SNP Analaysis of Alzheimer's Disease				
ABCA1	APOE	DSG2	MMP3	SORL1
ABCA7	BIN1	EPHA1	MS4A4E	SPON1
ACE	CD2AP	FERMT2	MS4A6A	TNF
APOC1	CD33	GRIN2B	NOS3	TOMM40
APOC2	CLU	HFE	PICALM	TRIP4
APOC4	CR1	MEF2C	PVRL2	ZCWPW1

Table 2. Genes identified through the GWAS catalogue, and prioritized using the ENDEAVOR pathway analysis tool. SNPs on genes were then filtered using Fst and D Jost to determine which showed the greatest differentiation between controls and AD.

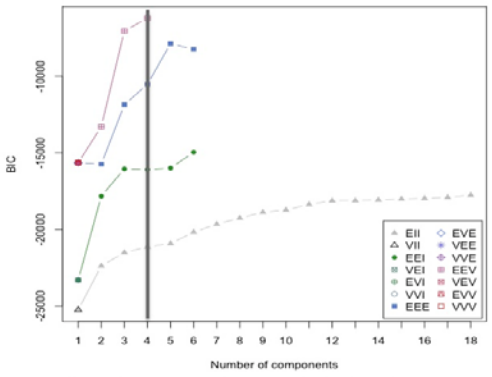


Figure 1. We used Bayesian Information Criterion (BIC) to determine the ideal number of clusters in our data. The vertical gray bar indicates an inflection point that was used to make this decision.

Cluster Groups				
	Cluster 1	Cluster 2	Cluster 3	Cluster 4
N	919	587	593	272
Age	78.66	78.11	76.01	80.32
Sex (% Female)	49.7%	52.8%	59%	*Too much missingness*
MMSE	25.02	25.58	24.63	*Too much missingness*
Braak staging	4.25	4.02	3.91	5
Cluster defining SNPs	rs2066715	rs2066715 rs2297398 rs4984338 rs207964 rs429358 rs7412	rs7588033 rs12610605 rs429358 rs7412	rs7588033 rs12610605 rs429358 rs7412

Table 1. Cluster information, including demographics, cognition, Braak & Braak pathology scoring, and the most distinguishing SNPs between clusters.

Disclosures: B.C. Riedel: None. **N. Jahanshad:** None. **D.A. Bennett:** None. **P.M. Thompson:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.13/W12

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA Grant U54 AG054345-01

Title: Molecular profiling of brain and retina to understand earliest stages of Alzheimer's disease

Authors: *S. R. CHINTALAPUDI[1], A. UYAR[2], H. WILLIAMS[1], X. WANG[3], G. CARTER[1], G. R. HOWELL[1]
[1]The Jackson Lab., Bar Harbor, ME; [2]The Jackson Lab., Farmington, CT; [3]Eisai Andover Innovative Medicines (AiM) Inst., Andover, MA

Abstract: Alzheimer’s disease (AD) is the most common form of dementia in the world. Lack of understanding of its prodromal phase precludes us effectively designing therapeutic interventions to target AD-vulnerable brain regions. Therefore, there is a dire need for the identification of novel molecular triggers that could serve as early markers and possible therapeutic targets for the disease. While AD-related pathology in the brain is well documented, the disease has also been reported to affect the retina, a developmental outgrowth of the brain. Our objective is to determine whether the retina can be used to ‘stage’ AD prior to significant development of AD in the brain. Our approach combines high-throughput transcriptomics with classical histological assessments and functional testing to identify novel molecular disease signatures during early disease stages in neural tissue initially, using *APP/PS1* mice, a commonly used model relevant to early-onset AD (EOAD). In the future, we plan to expand this approach to include additional existing EOAD models and new models of late-onset AD (LOAD) we are creating through Model Organisms Development and Evaluation for LOAD (MODEL-AD), a NIA-funded collaboration between The Jackson Laboratory, Indiana University and Sage Bionetworks.
To determine the earliest processes occurring prior to, and at the onset of, plaque deposition we performed gene expression profiling by RNA sequencing (RNA-seq) of brains and retinas from female *APP/PS1* mice and C57BL/6J (WT) controls at 2, 4, 5, and 6 months of age (prior to and during the onset of plaque deposition in the *APP/PS1* mouse). Differential expression analysis identified activation of the Janus kinase/signal transducer and activator of transcription (JAK/STAT) pathway and long-term potentiation as early events in the brain, in agreement with other published studies. Our analyses have identified phosphorylation of STAT3 in multiple cell types including astrocytes, microglia and neurons suggesting multiple roles for the JAK/STAT pathway in this model.
To explore the potential of the retina as a biomarker in AD, gene set enrichment using STRING identified vascular dysfunction as one of the critical early events in both the eye and the brain

which is being validated using a variety of *in-vivo* and histological approaches. Elucidation of vascular dysfunction as a marker and possible early driver for AD in both these tissues could represent a significant advance in understanding the biological underpinnings of AD and at the same time it may facilitate the identification of causal proteins and pathways for novel therapeutic approaches.

Disclosures: S.R. Chintalapudi: None. **A. Uyar:** None. **H. Williams:** None. **X. Wang:** None. **G. Carter:** None. **G.R. Howell:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.14/W13

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: The National Science Foundation of China, 81471313 and 91649119

Title: TET3-mediated DNA demethylation regulates AD neurodegeneration

Authors: *J. LI
Kunming Inst. of Zoology, Chinese Acad. Scie, Yunnan, China

Abstract: AD neurodegeneration has been associated with abnormal alterations in multiple epigenetic systems including aberrant DNA methylation/demethylation patterns. DNA 5-Hydroxymethylcytosin (5-hmC), a new epigenetic hallmark which was discovered recently, is becoming an interesting topic in brain ageing and age-related neurodegenerative disorders. However, the exact function of this base remains being poorly elucidated.Here, we found thatDNA dioxygenaseTET3-mediated conversion from DNA 5-methylcytosine (5-mC) to 5-hmC was involved in AD neurodegeneration.In both mouse and human ADbrain, a significant loss of 5-hmC was also found in specific brain regions including PFC and CA1 but not cerebellum in postmortem samples from patients with confirmed diagnosis of AD in comparison to age- and sex-matched healthy control subjects. Genome-wide analysis of 5hmC-seq data shows a significant shift in 5hmC enrichment in both regulatory and elements and repeated regions in AD neurons. Aβ directly attenuates TET3-mediated 5hmC production. Manipulation of TET3 activity affects the degenerative process of AD neurons.These results indicate that the involvement of 5-hmC in the pathology of neurodegenerative disorders and points out common and particular signatures that might aid in differential diagnosis as biomarkers or in the discovery of novel therapeutic targets.

Disclosures: J. Li: A. Employment/Salary (full or part-time):; Kunming Institute of Zoology, CAS.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.15/W14

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH AG034214

Title: Apoe locus dna methylation in alzheimers disease

Authors: Y. SHAO[1], G. D'ALEO[1], M. KHRESTIAN[1], J. ZAHRATKA[1], S. RAO[2], J. PILLAI[2], J. LEVERENZ[2], *L. BEKRIS[3]
[1]Genomic Med., [2]Luo Ruvo Ctr. for Brain Hlth., Cleveland Clin., Cleveland, OH; [3]Cleveland Clin. Genomic Med. Inst., Cleveland, OH

Abstract: In humans and in mouse models, *APOE* ε2 carriers have higher apoE levels, compared to ε3 and ε4 (ε2>ε3>ε4), and are protected against Alzheimer's disease (AD) pathology, such as the toxic Aβ peptide, suggesting that *APOE* ε2 plays an important role in the central nervous system. It has been suggested that if apoE2 or apoE3 protein levels are therapeutically increased, without increasing apoE4, this would allow for better clearance and less deposition of the toxic Aβ in amyloid plaques in AD brain. Many questions still remain on how *APOE* expression is regulated by distal elements at this locus and how they might be exploited to modulate apoE levels in AD. We have described the underlying mechanistic influence by *APOE* exon 4 regulatory element at the *APOE* ε2/ε3/ε4 haplotype location. Notably, the activity of this exon 4 regulatory element appears to depend on epigenetic factors that either enhance transcription or decrease expression depending on the *APOE* ε2/ε3/ε4 haplotype. The central hypothesis of this investigation is that the complex regulatory structure surrounding the *APOE* gene is regulated by key epigenetic factors depending on tissue type and disease status. DNA was extracted from human brain (n=12) and whole blood (n=85) and percent methylation was measured using the EZ DNA Methylation™ Kit (Zymo Research) and Infinium HumanMethylation450 BeadChip Kit (Illumina). Percent methylation of each cg site was evaluated for an association with disease status and age using a multivariate analyses (SPSS). Extended methylation analysis of the *APOE* locus suggests that methylation at *TOMM40* and *APOC1*, but not the *APOE* exon 4-3'UTR CpG, are different between AD and controls in the hippocampus. Methylation at the *APOE* exon 4-3'UTR CpG island, *APOC1* and *APOC1P1* is different in MCI compared to controls in whole blood. This exploratory DNA methylation analysis of an extended region of the *APOE* locus identified regions outside of the *APOE* gene that are differentially regulated according to disease status. These results suggest that regions surrounding the *APOE* gene, in addition to *APOE*, are differentially methylated according to disease status, and identifies regions that may be regulated by methylation at this locus.

Disclosures: **Y. Shao:** None. **G. D'Aleo:** None. **M. khrestian:** None. **J. Zahratka:** None. **S. Rao:** None. **J. Pillai:** None. **J. Leverenz:** None. **L. Bekris:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.16/W15

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: PRKDC and alzheimer's disease: A candidate gene study

Authors: *J. L. WEBB
Iowa State Univ., Ames, IA

Abstract: Background: A DNA-dependent protein kinase responsible for repairing non-homologous DNA strand breakage is encoded by the *PRKDC* gene. DNA repair deficiency is hypothesized to predispose individuals to neurodegenerative disease by affecting reactive oxygen species that damage neurons. The effects of mutations in genes regulating DNA repair such as PRKDC on clinical, cognitive and neurobiological indices have yet to be characterized in humans. This candidate gene study examined how PRKDC variation influences outcomes across the Alzheimer's disease (AD) spectrum using gene based and single variant association analysis. **Methods:** 757 non-Hispanic Caucasian participants from the AD Neuroimaging Initiative (ADNI) cohort underwent whole genome sequencing, where 404 of those subjects underwent T1-weighted structural MRI scanning. Genomic data was imputed from Illumina Human610-Quad BeadChip data and exported in SPSS 24 to assess main effects of genomic variants on neuroimaging, plasma and cerebrospinal fluid (CSF) biomarkers. FreeSurfer 6.0 measured cortical thickness and surface area of neuroanatomical structures. Covariates included age, gender, education, and baseline diagnosis. **Results:** 1239 variants were extracted from a region ±2 kb from PRKDC. Logistic regression models highlighted that subjects carrying the minor allele at rs10097783 were at a 27% increased risk of developing AD (Wald=4.211, P=0.041). Minor allele carriers also displayed lower CSF $A\beta_{1\text{-}42}$ ($F=3.99$, $P = 0.032$), MMSE score ($F=6.01$, $P < 0.01$), and increased insulin resistance ($F=5.66$, $P < 0.01$). Whole gene analysis indicated that the PRKDC gene is associated with bilateral entorhinal cortex surface area. **Conclusions:** These results suggest that PRKDC may affect AD risk by modulating putative peripheral and central biomarkers, highlighting the need to validate these results in independent, larger samples. Overall these results demonstrate that neuroinformatic approaches may identify additional genetic variation explaining unaccounted heritability in AD.

Disclosures: **J.L. Webb:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.17/W16

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: SYNAPSIS Foundation

Béatrice Ederer-Weber Stiftung

Alzheimer’s Association (NIRG-15-363964

Title: PM20D1 methylation quantitative trait locus is associated with Alzheimer’s disease

Authors: *J. SANCHEZ-MUT[1], H. HEYN[2], P. GARCIA-ESPARCIA[3], E. VIDAL[4], S. SAYOLS[5], L. GLAUSER[1], A. MONTEAGUDO-SÁNCHEZ[6], J. PEREZ-TUR[7], I. FERRER[3], D. MONK[6], M. ESTELLER[6], J. GRÄFF[1]
[1]BMI, EPFL, Lausanne, Switzerland; [2]CNAG, Barcelona, Spain; [3]Univ. of Barcelona, Barcelona, Spain; [4]CRG, Barcelona, Spain; [5]Inst. of Mol. Biol., Mainz, Germany; [6]IDIBELL, Barcelona, Spain; [7]Inst. de Biomedicina de València-CSIC, Valencia, Spain

Abstract: The chances to develop Alzheimer’s disease (AD) result from a combination of genetic and non-genetic risk factors, the latter likely mediated by epigenetic mechanisms. Genetic risk factors are usually interrogated by genome-wide association studies (GWAS), which have identified an important number of loci associated with AD pathology in the past. However, GWAS tend to result in strong statistical associations of only large portions of the genome, for which a causal relationship is difficult to establish. In contrast, locus-specific or epigenome-wide association studies (EWAS) identify site-specific epigenetic alterations providing mechanistic insights for a particular risk gene, but lack the statistical significance of GWAS. Combining both approaches, it has recently become possible to identify single nucleotide polymorphisms (SNPs) that correlate with alterations in DNA methylation levels - so-called methylation quantitative trait loci (mQTLs) - the importance of which for complex diseases has just started to be recognized. mQTLs have been reported for several neurological disorders including schizophrenia, obsessive-compulsive and bipolar disorder, but, thus far, not for neurodegenerative diseases such as AD.
Here, we report a hitherto unidentified association of the Peptidase M20 domain-containing protein 1 (PM20D1) with Alzheimer’s disease (AD). We find that PM20D1 is a methylation/expression quantitative trait locus (mQTL/eQTL) coupled to an AD-risk associated haplotype, which displays enhancer-like characteristics and contacts the PM20D1 promoter via a CTCF-mediated chromatin loop. PM20D1 is increased following AD-related neurotoxic insults, at symptomatic stages in the APP/PS1 mouse model of AD and in human AD non-risk haplotype

carriers, while its overexpression reduces oxidative stress-induced cell death and amyloid-β levels.

Disclosures: **J. Sanchez-Mut:** None. **H. Heyn:** None. **P. Garcia-Esparcia:** None. **E. Vidal:** None. **S. Sayols:** None. **L. Glauser:** None. **A. Monteagudo-Sánchez:** None. **J. Perez-Tur:** None. **I. Ferrer:** None. **D. Monk:** None. **M. Esteller:** None. **J. Gräff:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.18/W17

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH AG046200

Alzheimer Association IIRG 10-173180

MUSC interprofession

Title: Retinal dysfunction and behavior in an Alzheimer mouse model may be modified by diet

Authors: ***K. SAMBAMURTI**[1], P. VASUDEVARAJU[2], E. AMELLA[3], M. A. PAPPOLLA[4], C. SABIN[5], N. H. GREIG[6], D. K. LAHIRI[7]
[1]Neurosciences, MUSC, Charleston, SC; [2]Neurosciences, Padmaraju Vasudevaraju Ph. D., Charleston, SC; [3]Col. of Nursing, Elaine Amella, Charleston, SC; [4]Neurol., Univ. of Texas, Galveston, TX; [5]Neurosciences, Christina Sabin, Charleston, SC; [6]Drug Design & Develop. Section, LNS, Intramural Res. Program, Natl. Inst. On Aging, NIH, Baltimore, MD; [7]Dept. of Psychiatry, Indiana Univ. Sch. of Med., Indianapolis, IN

Abstract: Alzheimer disease (AD) and several retinopathies, including Age related macular degeneration (AMD) are linked to protein accumulation, in particular the Alzheimer amyloid beta protein (Aβ) and the microtubule associated protein Tau (MAPT). both been identified as major risk factors for AD and AMD. Interestingly, the amyloid β protein of 42 aa (Aβ42) is deposited in extracellular lesions in both these diseases. A number of studies have focused on reducing Aβ42 for treating AD. Critical to the understanding of AD is the recognition that Aβ accumulation does not take place in most individuals. Homocysteine and cholesterol were both identified as important risk factors liked to increases in AD pathology and dementia. Homocystinuria, a disease linked to failure of methionine catabolism leads to neurodegeneration with poor brain development and retinal function. The only treatment available to patients is restriction of methionine to prevent homocysteine accumulation. Lifelong restriction of methionine in diets preserves function in children with homocystinuria. Mice expressing FAD mutant APP or genomic human MAPT show retinal dysfunction as determined by

electroretinograms (ERGs). We find that restricting methionine in cell cultures drastically modifies Aβ protein precursor (APP) processing to reduce levels of its C-terminal fragments (CTFs) generated by α- and β-secretases, and also reduces the yield of Aβ. Moreover, a diet restricting methionine while maintaining calorie and nitrogen content reduces APP-CTFs and Aβ in an AD transgenic mouse model. In addition to APP processing, the post-transcriptional regulation of APP cannot be ruled out. Indeed, we are testing the effects of the treatment on microRNAs (miRNA) that specifically target APP and BACE1 protein expression. In summary, the treatment in our conditions improves retinal function, as measured by ERG and also corrects behavior deficits as measured by novel object preference after methionine restriction. We expect that this type of dietary restriction can be readily incorporated in persons at risk to prevent AD and AMD.

Disclosures: K. Sambamurti: None. **P. Vasudevaraju:** None. **E. Amella:** None. **M.A. Pappolla:** None. **C. Sabin:** None. **N.H. Greig:** None. **D.K. Lahiri:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.19/W18

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant AG051086

Title: Specific MicroRNAs regulating expression of proteins involved in Alzheimer's disease

Authors: *D. K. LAHIRI[1], N. CHOPRA[1], B. L. BAYON[2], N. H. GREIG[3], K. SAMBAMURTI[4]
[1]Dept. of Psychiatry, [2]Dept. of Med. & Mol. Genet., Indiana Univ. Sch. of Med., Indianapolis, IN; [3]Intramural Res. Program, Natl. Inst. On Aging, NIH, Baltimore, MD; [4]Dept. of Neurosciences, MUSC, Charleston, SC

Abstract: Small non-coding RNAs, in particular, have been the focus of many studies and their functional role in neurodegenerative diseases is currently being investigated. MicroRNAs (miRNAs) are non-coding small RNAs that are regulators of gene expression (post-transcriptionl) and consequently of many cellular processes. But their expression is often dysregulated in human diseases, including Alzheimer's disease (AD). Recently miRNAs were discovered in body fluids (serum, plasma) and brain tissues and their levels have often been reported to be altered in patients. Here we present evidence to show how particular miRNA species regulate neuronal genes involved in AD using neuronal cultures and human brain tissue specimens from control and AD subjects. AD results, in part, from over-production of amyloid-β peptide (Aβ), a proteolytic product of Aβ precursor protein (APP). Expression studies suggest

that dysregulation of proteins involved in Aβ production, such as APP and β-secretase, or BACE1, and/or Aβ degradation, such as neprilysin (NEP), contribute to excess Aβ deposition. Elucidating the regulation of these proteins' expression would ultimately reveal new drug targets. We report the regulation of these gene products by distinct miRNAs. Our results reveal a novel regulatory interaction between important AD-related genes (APP, BACE1 and NEP) and specific endogenously-expressed miRNA species. We also observed specific miRNAs (miR-101, miR-20b miR-153 and miR-346) regulating APP levels via interactions with the APP-3'UTR or APP-5'UTR in human primary neuronal cultures. These regulatory interactions may serve as novel therapeutic targets and enable the development of treatment strategies beneficial for AD.

Disclosures: D.K. Lahiri: None. **N. Chopra:** None. **B.L. Bayon:** None. **N.H. Greig:** None. **K. Sambamurti:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.20/W19

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: The role of Sp1-modulating compounds in Alzheimer's disease

Authors: *B. L. BAYON[1], B. MALONEY[2], X.-M. XU[3], R. R. RATAN[4], D. K. LAHIRI[2]
[1]Dept. of Med. & Mol. Genet., [2]Dept. of Psychiatry, [3]Program of Neurosci., Indiana Univ. Sch. of Med., Indianapolis, IN; [4]Burke-Cornell Med. Resch Inst., White Plains, NY

Abstract: Alzheimer's disease (AD) is characterized by extracellular amyloid-beta deposition. BACE1 is the beta-secretase responsible for the rate-limiting cleavage of amyloid precursor protein (APP) to amyloid-beta (Aβ). Sp1 binding sites on both the APP and BACE1 promoters implicate its potential role in AD. We demonstrate how these perturbations in Sp1 and other TFs may differ in activation in mammalian cells treated with Sp1 modulating drugs. To test the involvement of Sp1 in the regulation of APP, we treated several mammalian cells lines and post-mitotic human neuronal cells with mithramycin A (MTM), MTM analogs (MTM-SDK, MTM-SK), and tolfenamic acid (TA). Real time imaging was used to record changes in the neurobiology of cells during treatments. Sp1 inhibition via drug treatment or via siRNA transfection yielded minimal changes in confluence, cytotoxicity, neurite length, and neurite outgrowth. Treatment with MTM led to a significant decrease in APP and BACE1 expression and Aβ40 levels. Treatment with TA did not change Aβ40 levels or APP and BACE1 expression. Combination treatment of MTM and TA or of Sp1 siRNA transfection in TA treated cells led to high cytotoxicity. Our results suggest that when treated with MTM, a compensatory mechanism allows for neuronal cell survival perhaps via other members of the Sp family or other TFs. We also show that at non-toxic doses, MTM can decrease Aβ40 levels in human cells. Targeting TF

activity is a novel means to manipulate the amyloid pathway. Compounds modifying Sp1 and other TF binding to sites on the BACE1 and APP promoters may provide a means to limit the production of amyloid-beta and slow the symptoms of AD.

Disclosures: B.L. Bayon: None. **B. Maloney:** None. **X. Xu:** None. **R.R. Ratan:** None. **D.K. Lahiri:** None.

Poster

386. Alzheimer's Disease: Genetics and Functional Genomics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 386.21/W20

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: National Institute on Aging K01AG000986 (YJ)

National Institute on Aging P30AG028383 (YJ)

Virginia Center on Aging (RHL)

Inova Research Foundation (RHL)

George Mason University Research Foundation (ML)

Title: GRIN2B promoter polymorphism and processing speed in older adults

Authors: *R. H. LIPSKY[1,2], M. LIN[2], G. JICHA[3], X. DING[4], S. L. MCILWRATH[3], D. FARDO[3], L. BROSTER[3], F. SCHMITT[3], R. KRYSCIO[3], Y. JIANG[3]
[1]Neurosciences Res., Inova Hlth. Syst., Falls Church, VA; [2]George Mason Univ., Fairfax, VA; [3]Univ. of Kentucky Chandler Med. Ctr., Lexington, KY; [4]Western Kentucky Univ., Bowling Green, KY

Abstract: The genetic basis for inter-individual differences in mental processing speed with aging is not well understood**.** Animal studies provided a link between enhanced N-methyl-D-aspartate receptor function, particularly involving the GluN2B subunit encoded by the GRIN2B gene, and increased memory performance. We investigated the role of a single nucleotide polymorphism rs3764030 (G>A) within the human GRIN2B gene promoter in mental processing speed in healthy, cognitively intact, older adults. The A allele predicted creation of an E26 transcription factor family binding site. We performed in vitro DNA-binding and reporter gene expression assays of different rs2764030 allele combinations in transfected cells to show that the A allele was a gain-of-function variant, increasing GRIN2B mRNA levels. We tested the hypothesis that the A allele would be associated with better memory performance, e.g. faster reaction times during working memory. Twenty-eight older adults (ages 65-86) were recruited

and performed a visual working memory task. The rs3764030 polymorphism was genotyped and participants were grouped based on the presence of the A allele into GG and AA/AG. Carriers of the A allele maintained their processing speed over age compared to GG carriers (p= 0.026 slope of the regression line between AA and AG versus GG groups). To validate the results, 16 older adults from the same cohort participated in a different version of the short-term memory task. We found that reaction times were significantly slower with age in older adults with G allele ($p < 0.001$). Together, these findings support a role for rs3764030 in memory performance.

Disclosures: R.H. Lipsky: None. **M. Lin:** None. **G. Jicha:** None. **X. Ding:** None. **S.L. McIlwrath:** None. **D. Fardo:** None. **L. Broster:** None. **F. Schmitt:** None. **R. Kryscio:** None. **Y. Jiang:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.01/W21

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: MIUR, PRIN 2015, 2015N4FKJ4

Title: Molecular links of DNA replication to apoptosis in ß-amyloid-treated neurons

Authors: F. CARACI[1], R. SANTANGELO[1], A. MUNAFÒ[1], F. NICOLETTI[2], *A. G. COPANI[1]
[1]Univ. of Catania, Dept. Drug Sci., Catania, Italy; [2]Univ. Sapienza, Roma, Italy

Abstract: Expression of cell cycle proteins and replicative DNA synthesis have been observed in neuronal populations that eventually degenerate in the Alzheimer’s disease (AD) brain (Herrup et al., J Neurosci 24, 2004). In cultured rodent neurons, synthetic ß-Amyloid (Aß) reproduces the neuronal cell cycle re-entry observed in the human AD brain, and blockade of cell cycle activation prevents Aß-induced neurodegeneration (Copani et al., J Neurosci 26, 2006). DNA polymerase-ß (DNA pol-ß) plays a causal role in the DNA replication process that contributes to generate a death signal in neurons (Copani et al., J Neurosci 26, 2006; Merlo et al., J Nat Prod 78, 2015). Analysis of the molecular mechanism(s) linking DNA replication to neuronal apoptosis might lead to the identification of new neuroprotective strategies. In pure rat neuronal cultures challenged with synthetic Aß(1-42) oligomers, we are examining the role of claspin, a protein required for the activation of the conserved checkpoint pathway started by DNA replication stress (Lee et al., *J Mol Cell*. 11, 2003). On cross-linked nucleoprotein fragments from Aß-treated neurons, we found that claspin co-immunoprecipitated with cell division cycle 45 (Cdc45) at early times following Aß treatments, and disappeared at times coinciding with neuronal apoptosis (as assessed by cytofluorimetric analysis). We also found that

selective inhibitors of caspase-7, an enzyme known to promote claspin inactivation, kept neurons in S phase and reduced apoptosis. These data suggest that claspin and the downstream checkpoint signaling kinase, Chk1, might constitute the molecular link between DNA replication and apoptosis in Aß-treated neurons.

Disclosures: **F. Caraci:** None. **R. Santangelo:** None. **A. Munafò:** None. **F. Nicoletti:** None. **A.G. Copani:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.02/W22

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: funding from Mitsui Sumitomo Insurance Welfare Foundation Research Grant 2016

Title: AC253 improved acute cognitive deficits caused of β-amyloid and human amylin delivered by an intracisternal injection in conscious mice

Authors: *R. KIMURA[1], M. ZAYASU[2], M. SAIKI[2], A. INOUE[3]
[1]Ctr. for Liberal Arts and Sci., [2]Dept. of Applied Chemistry, Fac. of Engin., Tokyo Univ. of Science, Yamaguchi, Sanyo-Onoda, Japan; [3]Dept. of Neurology, Fac. of Med., Kyoto Univ., Kyoto, Japan

Abstract: The amylin receptor is a potential target receptor for expression of the deleterious actions of soluble oligomeric β-amyloid peptide (Aβ) species, and AC253, a novel amylin receptor antagonist, neutralizes the depressant effects of Aβ and human amylin on hippocampal long-term potentiation (LTP) in mice brain slice experimentation. In addition, depressed levels of LTP observed in 6- to 12-month-old transgenic mice, which overexpress amyloid precursor protein (TgCRND8), is also restored with AC253 (Kimura et.al. J Neurosci 2012). Those show that the effects of Aβ and human amylin on LTP are expressed via the amylin receptor, and blockade of this receptor by AC253 increases LTP even in transgenic mice that show increased brain amyloid burden. In this study, our purpose is to elucidate whether acute care of AC253 improves cognitive deficits in a model mouse of Alzheimer's disease. In order to accurate delivery of these peptide quickly, we administered directly into the brain stem. By that means, we adopted a bent needle with special angle for intracisternal (i.cist.) injection into conscious mice (Ueda at.el. EUR J PHARMACOL 1979). To determine whether the peptide affects cognitive function, we performed Y-maze Spontaneous Alternation test at 30 min after i.cist, administration. First, for making sure of drug administration, we tested the effect of accute 0.01 ml Scopolamine (1 mM) or PBS administration on Y-maze test, and found the clearly difference in the levels of spontaneous alternation performance, but not number of arms entered. Next, we

investigate that 0.01 ml Aβ (1 mM) or human Amylin (1 mM) administration in mice. Both showed dramatically reduced levels of spontaneous alternation performance in the Y-maze as compared with 0.01 ml PBS administration as a control. Furthermore, another i.cist. administration of AC253 (5 mM) at 30 min before Aβ or human Amylin administration recovered from the depressant effects on spontaneous alternation performance in the Y-maze. Our next step should be to examine whether depressed levels of spontaneous alternation performance on Y-maze test observed in transgenic mice, which overexpress amyloid precursor protein (5XFAD), could be restored with AC253. Then we show that human Amylin receptor antagonists serve as potentially useful therapeutic agents in severe level of Alzheimer's disease.

Disclosures: **R. Kimura:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Mitsui Sumitomo Insurance Welfare Foundation Research Grant 2016. **M. zayasu:** None. **M. saiki:** None. **A. inoue:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.03/W23

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Live cell integrated surface plasmon resonance sensing system for monitoring and electrical modulation of neuron signaling

Authors: *M. MOZNEB, A. NILCHIAN, C.-Z. LI
Biomed. Engin., Florida Intl. Univ., Miami, FL

Abstract: Being the 6th leading cause of death in the United States, Alzheimer's Disease (AD) is the most common form of dementia, in which symptoms gradually worsen over a number of years. It is believed that damage to the brain starts a decade or more prior to surfacing symptoms and cognition loss, therefore early detection of universal biomarkers is essential in diagnosis and treatment of the disease. Amyloid, Tau and Cholinergic hypotheses are the leading thoughts on which most currently available drug therapies exist. Nevertheless, the major cause of AD is yet unknown. Due to lack of comprehensive human cellular models, it has been a challenge to study the disease under all its physiological factors. A compatible cellular model would ideally include the effect of electrical and magnetics brainwaves as well as its chemical interactions and cellular communications. Our recent studies have been on evaluating different strategies to identify the fastest and most efficient way of immobilizing and orienting Immunoglobulin G (IgG) antibody. Using electrochemical surface plasmon resonance (EC-SPR), we have established and formulated protocols to orient antibodies under specific electrical charge for higher affinity of

interaction and better detection. Developmental studies were carried out on several trials for voltage, time and pH optimization of the immobilized IgGs. The results suggest that applying positive voltage to immobilized IgGs will give a higher affinity to antibodies for influenza peptide detection, than any other chemical immobilization methods. Our current research is focused on developing a 3D SPR based model for studying the relationship of Amyloid beta (Aβ) oligomers with neuron signaling under electrically controlled and stimulated environment. Human neurons will be grown inside of the SPR flow cell gasket and will be exposed to physiological amounts of Aβ peptides. The accumulation of Aβ and cell signaling will be monitored over time using EC-SPR while the system is being exposed to different electrical signals and electrical fields of varying strengths. We hypothesize that our model will provide better understanding of the effect of Aβ oligomers on neuron signaling and its related neurotransmitter release.

Disclosures: **M. Mozneb:** None. **A. Nilchian:** None. **C. Li:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.04/W24

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: The influence of β1-42-amyloid and cyclosporine A on the calcium channels of hippocampal neurons

Authors: *I. KRAVENSKA, V. YAVORSKY, I. MELNICK, O. LUKYANETZ
Bogomoletz Inst. of Physiol. of NASU, Kyiv, Ukraine

Abstract: The disorders of cellular calcium homeostasis may play an important role in the development of Alzheimer's disease. The aim of our studies was to establish the role of membrane calcium channels of neurons and the mitochondrial permeability transition pores in the development of Alzheimer's disease. The study was performed on rat hippocampal cultured neurons. The model of Alzheimer's disease was realized using incubation of cells with β1-42-amyloid (2 μM, 24 h). The contribution of mitochondrial pores in the studied process was evaluated by the effect of their specific inhibitor cyclosporine A (1.25 μM). It was used a patch-clamp ("whole cell") for calcium current measurements. The densities of calcium currents per the capacity of cell membranes were compared. The results were obtained with 57 hippocampal neurons, selected by their pyramidal shape. We have shown that the densities of calcium currents in neurons were not significantly affected by β1-42-amyloid and cyclosporine A (9% and 35% of the cells compared to control respectively) because of wide range of value scattering. The amplitudes of the currents were also independent on the capacity of control cell membranes and those incubated with experimental agents. However, under control conditions the maximum of

voltage-current curve of calcium currents was observed at the test membrane depolarization from +5 to +15 mV (100% of the cells), while after incubation with β1-42-amyloid and cyclosporine A at 0 mV (17% and 13% of the cells respectively).Thus, according to the chosen cellular model of Alzheimer's disease we did not observe significant changes in the amplitudes and densities of membrane calcium currents in neurons after incubation with β-amyloid and cyclosporine A. Although in number of cells we observed the shift of voltage-current curves toward to the hyperpolarization after the treatment.

Disclosures: I. Kravenska: None. **V. Yavorsky:** None. **I. Melnick:** None. **O. Lukyanetz:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.05/W25

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NHMRC

Title: Investigating Aβ toxicity using differentiated neuronal human stem cell cultures

Authors: *M. S. TAN[1], R. CAPPAI[1], M. DOTTORI[2], G. D. CICCOTOSTO[1]
[1]Dept. of Pathology, [2]Ctr. for Neural Engin., The Univ. of Melbourne, Parkville, Australia

Abstract: Alzheimer's disease (AD) is a neurodegenerative disease that primarily affects individuals aged 65 years and over. It is pathologically characterized by abnormal deposition of extracellular amyloid plaques and intraneuronal neurofibrillary tangles, progressive brain atrophy, and gradual synaptic and neuronal losses. Although the aetiology of AD remains elusive, studies have shown that amyloid beta (Aβ), a cleavage product from amyloid precursor protein (APP), is a key protein causing AD. More recent studies have shown that soluble low molecular weight Aβ oligomers correlate better with synaptic losses and are a better predictor of disease than amyloid plaque or neurofibrillary tangles (Sokolow, 2012). In the brain of individuals with AD, different neuronal subpopulations appear to exhibit vulnerability to Aβ; particularly the basal forebrain cholinergic and hippocampal glutamatergic neurons, while GABAergic neurons appear to remain relatively unaffected till later disease stages. Current treatments targeting cholinergic and glutamatergic systems only alleviate symptoms and are generally ineffective in halting disease progression.
Previous studies in our lab has shown a time and concentration dependent increase in Aβ binding that correlated with neurotoxicity in mouse cortical cultures (Jana, 2016). We hypothesize that Aβ exerts its neurotoxic effects by binding to a subpopulation of mature neurons. To better understand the mechanism(s) of Aβ-mediated toxicity on human neurons as well as human

neuronal subpopulation vulnerability, we generated human glutamatergic and GABAergic neurons using the human embryonic stem cell line H9. These neurons were cultured up to 12 weeks and treated with exogenously administered Aβ. We observed a concentration dependent increase in Aβ binding that correlated with neurotoxicity. Interestingly, we observed a trend of decreasing Aβ bound to cells at the later ages in both glutamatergic and GABAergic cultures. To identify the specific oligomeric Aβ species that is associated with cell death these differentiated neurons are being treated with purified Aβ oligomeric species (monomers, dimers, trimmers and tetramers) and correlated with toxicity.

Disclosures: M.S. Tan: None. **R. Cappai:** None. **M. Dottori:** None. **G.D. Ciccotosto:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.06/W26

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: UH Foundation

NIGMS-NIH, P20GM103451

NCGR

Title: Transcriptome profile of nicotinic receptor-linked beta amyloid neurotoxicity

Authors: *K. ARORA[1], M. BELCAID[2], R. A. NICHOLS[1]
[1]Cell and Mol. Biol., John A Burns Shool of Medicine, Univ. of Hawaii, Honolulu, HI; [2]Pacific Ctr. for Emerging Infectious Dis. Res., John A. Burns Sch. of Medicine, Univ. of Hawai'i, Honolulu, HI

Abstract: A better understanding of the global gene changes underlying the progression of Alzheimer's disease (AD) pathogenesis could help develop potential therapeutic strategies for the management of this significant neurodegenerative disorder. RNA sequencing (RNA-seq) technology is a powerful way to profile the transcriptome with great efficiency and higher accuracy. RNA-seq technology has been recently employed in various neurological disorders. In this study, we used RNA-seq technology to analyze global differential gene expression in the model neuroblastoma cell line NG108-15, transfected with or without α4β2 nAChRs, and treated or untreated with full-length beta amyloid ($A\beta_{1-42}$), to gain insights into the underlying mechanisms of beta amyloid-induced neurotoxicity. Cells were treated with media or 100nM of $A\beta_{1-42}$ for 3 days, followed by extraction and purification of their RNA. Multiplexed RNA-seq libraries were prepared from the cellular RNA and paired-end 100-bp sequencing was conducted using an Illumina HiSeq 2500 sequencer. Fold changes in the expression of transcripts (and

hence, genes) between untreated and Aβ-treated samples were determined. Ingenuity pathway analysis (IPA) was employed to determine the cellular pathways. Similar experiments were conducted to compare gene expression between receptor expressing and mock (no receptor) control cells, treated or untreated with $A\beta_{1-42}$.
Our results demonstrate that treatment with $A\beta_{1-42}$ in α4β2 nAChR-transfected cells differentially modulated the expression of 15 prominent genes as compared to that of untreated cells. Differentially modulated genes included Rcan3, Chrna4, Apba3, Irak-1, Rab11 and Kif1c. Most notably, chrna4 and rcan3 (Regulator of Calcineurin), were upregulated 3-fold and 2-fold, respectively. The differential gene expression identified by RNA-seq was confirmed by real time PCR. We further confirmed the change in the protein expression of these genes by western blot. Experiments are ongoing to delineate the role of these genes in mediating beta amyloid-induced neurotoxicity.
In conclusion, this is the first study to conduct an entire transcriptome profile of model nerve cells expressing a defined high-affinity target receptor, namely nAChR, for beta amyloid. Characterizing the genes associated with neuronal death after beta amyloid treatment will have significant impact on developing neuroprotective agents to reduce or prevent neuronal death in AD patients.

Disclosures: K. Arora: None. **M. Belcaid:** None. **R.A. Nichols:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.07/W27

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Fondecyt Grants 1130068

Fondecyt Grants 1150736

CONICYT Ph.D. fellowship 21130445

Title: Mediators released by astrocytes in response to AβOs increase neuronal oxidative tone and decrease nuclear distribution of pSerStat3

Authors: *Y. A. MUÑOZ[1], A. PAULA-LIMA[2], M. T. NUÑEZ[1]
[1]Dept. of Biol., [2]Inst. for Res. in Dent. Sci., Univ. of Chile, Santiago, Chile

Abstract: Amyloid-beta oligomers (AβOs) have been found in Alzheimer's disease (AD) brains and there is vast evidence that supports a role of AβOs, which would trigger synapse failure and memory impairment in animals. Astrocytes respond to AβOs through a process called reactive astrogliosis, which generates reactive oxygen/nitrogen species (ROS/RNS) and inflammatory

cytokines that affect adjacent neurons. Stat3 is a crucial transcription factor involved in maintenance and function of nervous system and its deregulation has been implicated, with strong but controversial data, in AD. Growth factors induce serine-727 phosphorylation and this modification is associated with modulation of transcriptional activity of Stat3. The main goal in this work is determine whether hippocampal neuronal Stat3 is affected by reactive astrocytes. **Methods**: Primary hippocampal neuron and astrocytes cultures were used. Changes in pSerStat3 distribution were detected by immunocytochemistry. The oxidative tone and ROS production were evaluated by redox cytochemistry and Hyper strategy. Protein and mRNA levels were determined by Western blotting and qPCR, respectively. **Results:** We found that AβOs induced no changes in protein, mRNA Stat3 levels and pSerStat3 cellular distribution, in neurons. However, AβOs treatment in mixed neuron-astrocytes cultures induced notorious neuronal pSerStat3 redistribution. Reactive astrocytes increased the expression of pro-inflammatory cytokines, and astrocyte-conditioned media treated with AβOs (ACM-AβOs) increased the neuronal oxidative tone which it was abolished the antioxidant treatment. ACM-AβOs induced a decrease of Stat3 target genes (survival response) and an increase of pro-apoptotic Bax/Bcl2 ratio. **Conclusion:** We propose that in hippocampal neurons, pSerStat3 is a sensor for stressor astrocyte-produced induced by AβOs activation. **Financial support:** This work was supported by CONICYT Ph.D. fellowship 21130445 to YM and Fondecyt Grants 1150736 to APL and 1130068 to MTN.

Disclosures: Y.A. Muñoz: None. **A. Paula-Lima:** None. **M.T. Nuñez:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.08/W28

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Alzheimer’s Disease Research Center (NIH P50AG005146)

BrightFocus Foundation A2015332

NIH S10OD016374

Title: Alzheimer's disease: Aβ toxic conformer has dynamic localization in the human brain before Aβ plaques form

Authors: Y. KAGEYAMA[1], O. PLETNIKOVA[2], G. L. RUDOW[2], K. IRIE[3], S. M. RESNICK[4], D. R. FOWLER[5], L. J. MARTIN[2], *J. C. TRONCOSO[2]
[1]Pathology, Div. of Neuropathology, Johns Hopkins Univ. Sch. of Med., Baltimore, MD;
[2]Pathology, Div. of Neuropathology, Johns Hopkins University, Sch. of Med., Baltimore, MD;

[3]Grad. Sch. of Agriculture, Div. of Food Sci. & Biotech., Kyoto Univ., Kyoto, Japan; [4]LBN, Natl. Inst. On Aging, Baltimore, MD; [5]Office of the Chief Med. Examiner, Baltimore, MD

Abstract: Amyloid beta (Aβ) is an abundant brain protein involved in synaptic physiology and also critical to the pathogenesis of Alzheimer's disease (AD). In this study, we examined the distribution of Aβ in the brain of young subjects free of AD lesions. We used the 11A1 antibody, targeting a toxic form of Aβ, and immunofluorescence to localize 11A1 immunoreactivity in the parietal cortex in 35 subjects 30-65 years of age. We observed 11A1 immunoreactivity already present in cortical neurons, astrocytes, neuropil and pericapillary spaces at 30 years of age. Approximately 85% of neurons have the Aβ immunoreactivity, which colocalizes with the lysosomal marker Cathepsin D, suggesting degradation of the peptides in these neurons. Around 75% of protoplasmic astrocytes also contain the Aβ immunoreactivity. The proportion of neurons and astrocytes containing the Aβ immunoreactivity remains stable with age. The Aβ immunoreactivity was also identified in nearly 30% of the perivascular space of capillaries during the fourth and fifth decades. The Aβ immunoreactivity was also present in the neuropil as 1-2 µm round particles. Imaging of these novel particles with vesicle markers showed colocalization limited to CD63, which was also detected in astrocytes but not in neurons. This finding suggests that astrocytes participate in the processing of Aβ. Notably, in the sixth decade, at the same time that the proportion of pericapillary spaces with the Aβ immunoreactivity declined to nearly 20%, the number of neuropil Aβ particles sharply increased. In summary, Aβ is normally present in various cell types and brain structures, and appears to be constitutively produced, degraded and intracellularly-extracellularly trafficked in the brain. This state is reflected by a stable presence of Aβ in the neuropil until age 50 years. However, around this age, this steady state of Aβ changes with a decrease of pericapillary Aβ and a concomitant increase in neuropil Aβ. Autopsy tissues were obtained in collaboration with the Human Brain Tissue Repository of the Lieber Institute, directed by Thomas M. Hyde, M.D., Ph.D. and Joel E. Kleinman, M.D., Ph.D.

Disclosures: Y. Kageyama: None. **O. Pletnikova:** None. **G.L. Rudow:** None. **K. Irie:** None. **S.M. Resnick:** None. **D.R. Fowler:** None. **L.J. Martin:** None. **J.C. Troncoso:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.09/W29

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: CSIR-12FYP Network Project (BEND BSC-0206).

Title: Subtle genomic DNA damage leads to increase in production of Amyloid-β(1-42) without inducing reactive oxygen species

Authors: *H. DAS, S. C. BISWAS
Cell Biol. and Physiol., CSIR-Indian Inst. of Chem. Biol., Kolkata, India

Abstract: Alzheimer's disease (AD) is a progressive neurodegenerative disorder which is also known as the most common form of dementia worldwide. The factors associated with aging like mitochondrial dysfunction, genomic instability, loss of proteostatis etc contribute to the progression of this disease. A sudden misbalance between the formation and degradation/ clearance of intracellular amyloid beta (Aβ) starts appearing way before the real manifestation of the disease. In this study the major objective was to correlate the chemistry of an aging cell in vitro by introducing a very subtle amount of DNA damage inside differentiated SHSY5Y cells, hypothesizing whether this could in turn increase intracellular Aβ level or not. Series of doses of Camptothecin (CPT) starting from 5 nM to 10 µM was treated in differentiated SH-SY5Y cells for 24 h to check cellular viability and it was observed that upto 500 nM dose of CPT there was no significant amount of cell death. A window of dose of CPT was selected (5 nM- 50 nM) for further studies to measure the intrinsic stress response of differentiated SH-SY5Y cells under that minimal level of stress. A sign of very subtle genomic DNA damage (increased expression of pH2AX), hyper fused mitochondria (by Mito tracker red staining),hyper polarization of mitochondrial membrane (TMRM intensity) was observed from 5 nM dose itself but there was no increased level of intracellular reactive oxygen species (by DCFDA fluorescence) production up to 50 nM CPT. Interestingly there was an increased expression of complex V of oxidative hosphorylation chain of mitochondria but the expression of other complexes (I-IV) were unaltered when the cells were treated with 5 nM dose of CPT for 24 h. There is also a change in the expression of Mfn2 and VDAC1 from 5 nM CPT itself. Along with that there was an increase in the intracellular Aβ (1-42) level from 5 nM dose of CPT. Taken together this data suggest that increase of intracellular Aβ (1-42) is a stress response of a neuronal cell which is under subtle genomic DNA damage. Thus we propose that age related subtle genomic DNA damage may lead to increased Aβ (1-42) production which may cause oxidative stress and further production of Aβ (1-42).

Acknowledgements:
The work is supported by CSIR-12FYP Network Project (BEND BSC-0206).

Disclosures: H. Das: None. **S.C. Biswas:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.10/W30

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Biochemical analysis of oxygenated Aβ by photooxygenation system

Authors: *Y. HORI[1], S. OZAWA[1], A. TANIGUCHI[2], Y. SHIMIZU[2], R. KINO[2], T. SAITO[3], T. C. SAIDO[3], Y. SOHMA[2], M. KANAI[2], T. TOMITA[1]
[1]Lab. of Neuropathology and Neurosci., Grad. Sch. Pharm. Sci., The Univ.Of Tokyo, Tokyo, Japan; [2]Lab. Synthetic Organic Chem., Grad. Sch. Pharm. Sci., The Univ. of Tokyo, Tokyo, Japan; [3]RIKEN Brain Sci. Inst., Saitama, Japan

Abstract: One of the pathological hallmarks of Alzheimer disease (AD) is a senile plaque, which is composed of aggregated amyloid-β peptides (Aβ). Several lines of evidence suggest that the aggregation process from monomer Aβ to its aggregates is associated with the neurotoxicity. Thus, inhibition of Aβ aggregation would provide therapeutic approaches against AD. Biochemical and cell biological studies indicate that oxygenated Aβ exhibits low aggregation potency and neurotoxicity than those of native Aβ, suggesting that artificial oxygenation of Aβ is a candidate for AD therapeutics. Thus, we synthesized a photooxygenation catalyst, which is activated by light and oxygenates Aβ (Taniguchi *et al*., Nat Chem 2016). This catalyst selectively oxygenated the synthetic aggregated Aβ and inhibited further aggregation and neurotoxicity. However, the effect of the catalyst on the aggregated Aβ *in vivo* remains unclear. To verify this issue, we carried out photooxygenation experiments of Aβ derived from AD model mice (NL-G-F mice, Saito et al., Nat Neurosci 2014) and analyzed by biochemical techniques. First, to check the photooxygenation activity for aggregated Aβ derived from the brains of AD model mice, the catalyst was mixed with the brain lysate and irradiated with 660 nm light. We confirmed the oxygenated Aβ peaks were detected using MALDI-TOF-MS. In addition, western blotting analysis showed the higher molecular weight bands (oxy-Aβ bands) than that of monomer Aβ by photooxygenation. Stepwise fractionation of photooxygenated brain lysate revealed that oxy-Aβ bands were detected in the insoluble fraction. These data indicated that catalyst has specifically oxygenated the aggregated Aβ in AD model mice brain. To further elucidate the effect of photooxygenation of Aβ in living animals, we constructed the repeated photooxygenation system for AD model mice. Catalyst was injected into hemi-hippocampus following the irradiation using LED fiber through the surgically inserted guide once in a day. Reactions were repeated for 7 times, and isolated hippocampi were analyzed. We observed the appearance of oxy-Aβ bands, indicating that oxygenation reaction for Aβ is occurred in living animal. Taken together, in this study, we revealed that photooxygenation catalyst oxygenates not only synthetic Aβ, but also aggregated Aβ derived from AD model mice. These data indicate therapeutic possibility of photooxygenation catalyst for AD.

Disclosures: Y. Hori: None. **S. Ozawa:** None. **A. Taniguchi:** None. **Y. Shimizu:** None. **R. Kino:** None. **T. Saito:** None. **T.C. Saido:** None. **Y. Sohma:** None. **M. Kanai:** None. **T. Tomita:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.11/W31

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant AG051266

BrightFocus Foundation

Alzheimer Association

Title: *In vivo* Aβ clearance from interstitial fluid and efflux through the choroid plexus are negatively influenced by peptide oligomerization

Authors: *A. A. ROSTAGNO, E. CABRERA, P. GIANNONI, F. MCINTEE, T. A. NEUBERT, J. GHISO
New York Univ. Sch. of Med., New York, NY

Abstract: Amyloid β (Aβ) is the major constituent of the brain deposits found in parenchymal plaques and cerebral blood vessels (CAA) of patients with Alzheimer's disease (AD). Several lines of investigation support the notion that synaptic pathology, one of the strongest correlates to cognitive impairment, is related to the progressive accumulation of neurotoxic Aβ oligomers. The process of oligomerization/fibrillization is concentration-dependent and is therefore highly reliant on the homeostatic mechanisms that regulate the steady state levels of Aβ influencing the delicate balance between rate of synthesis, dynamics of aggregation and clearance kinetics. Emerging new data suggest that reduced Aβ clearance, particularly in the aging brain, plays a critical role in the process of amyloid formation and AD pathogenesis. We have used a combination of intracerebral stereotaxic injections in C57BL/6 wild-type mice with biochemical and mass spectrometric analyses of CSF to evaluate the brain clearance and catabolism of well-defined monomeric and low molecular mass oligomeric assemblies derived from Aβ1-40 and Aβ1-42. Our data clearly demonstrate that Aβ physiologic removal from the brain is extremely fast, is negatively modulated by age, and involves local proteolytic degradation leading to the generation of heterogeneous C-terminally cleaved proteolytic products, while providing clear indication of the detrimental role of oligomerization for brain Aβ efflux. Immunofluorescence confocal microscopy studies provide insight into the cellular pathways involved in the brain removal and cellular uptake of Aβ. The findings indicate that clearance from brain interstitial fluid follows local and systemic paths and that in addition to the blood-brain barrier, local enzymatic degradation and the bulk flow transport through the choroid plexus into the CSF play significant roles. Our studies highlight the diverse factors influencing brain clearance and the participation of various routes of elimination opening up new research opportunities for the

understanding of altered mechanisms triggering AD pathology and for the potential design of combined therapeutic strategies.

Disclosures: **A.A. Rostagno:** None. **E. Cabrera:** None. **P. Giannoni:** None. **F. McIntee:** None. **T.A. Neubert:** None. **J. Ghiso:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.12/W32

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Fondecyt 1161078

Fondecyt 1130747

Title: Amyloid β-induced mitochondrial dysfunction is mediated by changes on PGC-1α and Mfn1/Drp1 on cellular models of Alzheimer Disease

Authors: ***J. FUENTEALBA**[1], J. PANES-FERNANDEZ[3], T. CELIS-MUÑOZ[2], T. SILVA-GRECCHI[2], D. MENNICKENT[2], P. A. GODOY[2]
[2]Physiol., [1]Univ. de Concepcion, Concepcion, Chile; [3]Physiol., Univ. of Concepcion, Concepcion, Chile

Abstract: Increased production of amyloid-beta (Aβ) peptide is one of the main features of Alzheimer's disease (AD). Soluble oligomers of Aβ (SOAβ) are postulated as the main toxic agent involved in the disease pathogenesis. Multiple toxic effects have been described in AD development, and could be replicated in different studies employing SOAβ: one of these features is mitochondrial dysfunction. The aim of this work was to evaluate the effects of SOAβ in two important mechanisms of mitochondrial function regulation, mitochondrial biogenesis and dynamics. Mitochondrial biogenesis is regulated by the key transcription regulator peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC-1α), which is in turn deacetylated and activated by the silent mating type information regulation 2 homolog (SIRT1) protein. On the other hand, mitochondrial dynamics are a highly regulated process involving fission and fusion events orchestrated by several proteins, including fusion protein Mitofusin 1 (Mfn1) and fission dynamin-related protein 1 (DRP1). Through western blot and immunofluorescence techniques, we observed decreased SIRT1 protein levels after SOAβ treatments (2h: 47±6%, 5h: 51±15%, 24h: 45±10%). onversely, an increment of PGC-1α protein was detected after 5h incubations (218±33%), though reduced levels were obtained after 1 and 24 h treatments (30±8%, 42±7% respectively). We studied the interaction between these two proteins after a 24 h SOAβ exposure, and curiously, the interaction was lost, which correlates

with a change in PGC-1α distribution, increasing its immunoreactivity in the cytoplasm (177±7%) and decreasing in the nucleus (38±3%). On the other hand, after 2 h incubations we observed a reduction in Mfn1 protein levels (42±11%), and an increase in DRP1 protein (198±27%); additionally, DRP1 presented a distribution shift on hippocampal neurons, increasing its immunoreactivity in neuronal processes. Changes regarding mitochondrial protein levels correlate with an alteration of mitochondrial network morphology, noted by fragmented mitochondrial populations in SOAβ-treated PC12 cells (< 2 μm, granular pattern), compared to control populations (2-6 μm, tubular pattern). Finally, these results suggest a SOAβ-induced imbalance in mitochondrial dynamics, through an early response mechanism to injury, represented by a fission potentiation and decreased mitochondrial biogenesis, thus blocking the pathway that lead to the cell to have functional mitochondrias and support the metabolic requeriments of the cell in the presence of SOAβ.

Disclosures: J. Fuentealba: None. **J. Panes-Fernandez:** None. **T. Celis-Muñoz:** None. **T. Silva-Grecchi:** None. **D. Mennickent:** None. **P.A. Godoy:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.13/W33

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH P30 NS47466

Title: Single D-peptide treatment causes local long-term changes in Aβ deposition in Tg AD model mice

Authors: *T. VAN GROEN[1], I. KADISH[1], N. JIANG[2], D. WILLBOLD[2]
[1]Cell, Developmental and Integrative Biol., Univ. Alabama-Birmingham, Birmingham, AL; [2]Forschungszentrum Jülich Gmbh, Jülich, Germany

Abstract: Our two transgenic mouse lines express two genes with AD mutations, i.e., PS1 (ΔE9 mutation) and APPswe or one gene with AD mutations APPswe/dutch/iowa. They develop plaques at about four months of age, and we have described that two types of deposits are present, plaques (with a thioflavine S positive core) and diffuse deposits. Furthermore, plaques are surrounded by activated glial cells, but diffuse deposits are not. We hypothesized that these distinct amyloid deposits would present differently following single intracerebral (hippocampus) fluorescently labeled anti Aβ42 oligomer targeted D-peptide injections in these mice.
We sacrificed and transcardially perfused the mice at two weeks and 3 months following the injections. The brains were cut and immunohistochemically stained for Aβ species, and GFAP and Iba-1.

Following the two week survival time the AD mice displayed a clear labeling of Aβ in the overlying cortex and hippocampus, and, locally a reduction in Aβ deposition was noted. No change was present in the number of labeled neurons. Further, the labeled Aβ is present in lysosomes in neurons near the injection site, but surprisingly no labeled Aβ is present in astrocytes or microglia. Labeled Aβ deposits seem to stay labeled over the researched timeframe, whereas the diffuse extracellular labeling (not bound to Aβ) disappears. Following the three months survival time the AD mice displayed a clear labeling of Aβ in the overlying cortex and hippocampus, and, locally a reduction in Aβ deposition was noted. It should be noted that none of these injections is associated with any glial activation, neither in the cortex nor in the hippocampus.
Thus, likely the D-peptides attach to the Aβ42 present in these amyloid deposits that develop in these AD model mice, and prevent the growth and buildup of these deposits.

Disclosures: T. Van Groen: None. **I. Kadish:** None. **N. Jiang:** None. **D. Willbold:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.14/W34

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: AG051521

AG050201

Title: Developmental Exposure to particulate air pollutants impairs hippocampal dependent memory, reduces hippocampal neurogenesis, and causes depressive behaviors

Authors: *N. C. WOODWARD[1], R. G. JOHNSON, III[2], F. SHIRMOHAMMADI[1], C. SIOUTAS[1], S. E. KANOSKI[1], C. E. FINCH[3], T. E. MORGAN[4]
[1]USC, Los Angeles, CA; [2]USC, Glendora, CA; [3]USC Col., [4]Davis Sch. of Gerontology, USC, Los Angeles, CA

Abstract: Traffic-related air pollution (TRAP) has increasing interest due to evidence linking increased risk of autism spectrum disorders to exposure to TRAP derived ambient particulate matter (PM) (Volk et al., 2013). This study examined the impact of nPM exposure during the critical window of development on longer term cognition. Sprague Dawley rats were exposed to nanoscale particulate matter (nPM, diameter < 200nm) from gestation to 28 weeks of age. nPM was collected adjacent to the CA-110 freeway in downtown Los Angeles, and is composed of the water-soluble fraction of ambient PM, composed primarily of organic carbons, nitrate, sulfate, ammonium. Pregnant rats were exposed 3 days/week, for 5 hours/day to 340 ug/m^3 nPM.

Developmental air pollution exposure impaired hippocampal dependent contextual memory, measured by novel object in context test. At six months of age, prenatally exposed rats spent less time exploring the novel object, resulting in a 20% reduction in the discrimination index, and a 60% reduced shift from baseline (investigation above chance levels). Exposed rats had a 30% decreased latency to first period of immobility, and spent 80% more total time immobile. nPM exposure also caused increased depressive behavior, measured by forced swim. Neurogenesis was impaired. The EDU assay of adult male hippocampal sections revealed decreased neurogenesis in the Cornu Ammonis 1 (CA1) and dentate gyrus (DG) by 60% or more, with no change in the CA3. The CA3 showed no effect of air pollution exposure in any subregion. Forced swim performance showed strong correlation with neurogenesis in, CA1, and DG. The behavioral deficits could be driven by an observed reduction in neurogenesis in the CA1 and DG of the dorsal hippocampus. The present data corroborates epidemiological findings of a developmental cognitive vulnerability to air pollution, and highlight the importance of reducing air pollution exposure in the population.

Disclosures: **N.C. Woodward:** None. **R.G. Johnson:** None. **F. Shirmohammadi:** None. **C. Sioutas:** None. **S.E. Kanoski:** None. **C.E. Finch:** None. **T.E. Morgan:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.15/W35

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R01AG051521, Finch PI

Title: Estradiol protective roles against air pollution mediated amyloidogenesis in neuroblastoma cells

Authors: *N. SAFI[1], ***N. SAFI**[1], M. CACCIOTTOLO[1], F. SHIRMOHAMMADI[2], C. SIOUTAS[2], T. MORGAN[1], C. FINCH[1]
[1]Leonard Davis Sch. of Gerontology, [2]Viterbi Sch. of Engin., USC, Los Angeles, CA

Abstract: Older women (particularly after menopause) are at higher risk of Alzheimer's Disease (AD), compared to men (Barnes et al; 2005). Physiological levels of 17-β-estradiol (E_2) can decrease the amyloid β-peptide (Aß) released by primary neuronal cultures derived from rodent and human cerebral cortex (Xu et al; 1998). However, the hormonal mechanism and the interaction with environmental factors during AD pathogenesis are not well understood (Janicki et al; 2010). Recent analysis of Women's Health Initiative Memory Study (WHIMS), showed that residence in polluted areas with elevated fine Particulate Matter ($PM_{2.5\mu m}$) had 92% higher risk of dementia (Cacciottolo et al; 2017). We hypothesized that E_2 may have a protective effect

against AD pathogenesis and air pollution particles. To test this, mouse neuroblastoma (N2a) cells expressing Swedish mutant APP (K595N/M596L) (N2a-APP/swe) were exposed to nano-sized sub fraction of $PM_{2.5\mu m}$ (nPM) (5 μg/ml) in presence and absence of 20 nM E_2 (physiological level). The toxicity was assessed by MTT and the transcriptional changes in APP, BACE1, and mERβ were analyzed by qPCR. In the MTT assay, nPM reduced mitochondrial activities of N2a/APPswe by 50% ($p<0.001$). Treatment with E_2 attenuated nPM effects on MTT by 30% ($P<0.001$). Similarly, the 2-fold nPM-induced increase of APP and BACE1 mRNA levels was attenuated by E2 ($p<0.0001$). In contrast mERβ was not induced by nPM, but was induced by E2. These findings suggest potential neuroprotection by E2 in resistance to air pollution PM.

Disclosures: N. Safi: None. **M. Cacciottolo:** None. **F. Shirmohammadi:** None. **C. Sioutas:** None. **T. Morgan:** None. **C. Finch:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.16/W36

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R21AG050201

R01AG051521

Title: Long term exposure to urban nano particulates has negative effects on cognitive functions and affects Alzheimer’s disease pathological hallmarks

Authors: *C. D'AGOSTINO[1], M. CACCIOTTOLO[1], F. SHIRMOHAMMADI[2], C. SIOUTAS[2], T. E. MORGAN[1], C. E. FINCH[1]
[1]USC Leonard Davis Sch. of Gerontology, Los Angeles, CA; [2]USC Viterbi Sch. of Engin., Los Angeles, CA

Abstract: In experimental rodent studies, chronic exposure to nano-sized particles from traffic-associated particulate matter (nPM- TRAP) robustly induced neuro- inflammatory responses (Morgan et al. 2011), amyloid accumulation and neurodegeneration (Cacciottolo et al., 2017). This study analyzed the impact of chronic nPM (< 200 nm) exposure on memory function and Alzheimer markers (Aβ38-40-42 accumulation) in wild type mice (C57BL/6J). Male mice aged 3 months were exposed to re-aereosolized nPM (350 $\mu g/m^3$) or to control filtered ambient air for 5 hours a day, 3 days per week, for 8 weeks. Memory was assessed by the ‘novel object recognition in contest (NOIC) test’ for hippocampal-mediated spatial recognition memory and learning. Mice exposed to nPM showed 25% deficit of memory compared to non-exposed

controls. Cerebral cortex lysates (PBS soluble supernatant, 100,000g x 60 min) were analyzed by ELISA for Aβ peptides. Exposed mice showed elevated soluble Aβ40 and -42 peptides in cerebral cortex by 1.7 and 2-fold vs controls. Our findings confirm studies of Fonken et al (2011) which showed impaired spatial learning from 9 months of TRAP exposure in the same mouse genotype, C57BL/6J males. Moreover, we found that the increase of Aβ was negatively correlated with memory function in nPM exposed mice, but not in control group. These findings suggest that endogenous rodent Aβ has synaptic activity relevant to cognitive functions. This association was not expected because rodent Aβ differs from the human in three amino acids that reduce its aggregation and toxicity (De Strooper et al 1995, Boyd-Kimball et al 2004). Thus, memory decline associated with TRAP in human populations may be due to increased levels of Aβ peptides, even in absence of detectable plaques.

Disclosures: C. D'agostino: None. **M. Cacciottolo:** None. **F. Shirmohammadi:** None. **C. Sioutas:** None. **T.E. Morgan:** None. **C.E. Finch:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.17/X1

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: AG051521

AG050201

Title: Developmental Exposure to particulate air pollutants causes neuroinflammation

Authors: *R. G. JOHNSON, III[1], N. C. WOODWARD[2], F. SHIRMOHAMMADI[3], C. SIOUTAS[3], S. E. KANOSKI[4], T. E. MORGAN[2], C. E. FINCH[2]
[2]Leonard Davis Sch. of Gerontology, [3]Viterbi Sch. of Engin., [4]Dornsife Col. of Letters, Arts and Sci., [1]USC, Los Angeles, CA

Abstract: Traffic-related air pollution (TRAP) has drawn interest due to evidence linking exposure to TRAP derived particulate matter (PM) and the risk of dementia (Cacciottolo2017) and autism-spectrum disorders (Volk 2015). This study focuses on nanoscale particulate matter, a subset of TRAP with a diameter of less than 200 µm. This study examined effect of nPM on exposure from before birth into adult life on systemic and neurodegenerative inflammatory markers. Sprague Dawley rats were exposed to nPM from gestation to 28 weeks of age. The nPM was collected adjacent to the CA-110 freeway in downtown Los Angeles. Rats were exposed on an alternate day intermittent schedule of three days per week, for five hours per day at 340 ug nPM/m3; controls were exposed to HEPA filtered air. Serum from nPM exposed rats at

the time of euthanasia had decreased levels of cytokines putatively associated with the M2 macrophage phenotype: IL-4, -20%; IL-10, -25%; IL-13, -15%. There was no change in serum levels of IL-6 and TNFα. In the hippocampus, microglial staining with Iba1 was increased by 40% in the dentate gyrus (DG) polymorphic layer, without change in the DG molecular layer. In the CA1 oriens layer, Iba1 staining was 30% higher in nPM animals vs controls, but with no difference in the CA1 radiatum. Also in the hippocampus, staining for the vascular endothelial cell maker (Zo-1) was reduced 75% in the CA1 oriens of the nPM exposed, with no difference in the CA1 radiatum.
These data show that exposure to nPM from early development into adult life may induce low grade systemic inflammation resulting from reductions in serum anti-inflammatory markers which could lead to increased neuroinflammation. Furthermore, the decrease in endothelial Zo-1 suggests alterations of the blood brain barrier. These changes may underlie some of the hippocampal-mediated behavioral deficits observed in this same study (Woodward et al).

Disclosures: R.G. Johnson: None. **N.C. Woodward:** None. **F. Shirmohammadi:** None. **C. Sioutas:** None. **S.E. Kanoski:** None. **T.E. Morgan:** None. **C.E. Finch:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.18/X2

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: the Cure Alzheimer’s Fund

R21AG040753

R01AG051521

R01AG050201

Title: Urban traffic-derived nanoparticulate matter promotes pro-amyloidogenic APP processing through increase of oxidative stress and lipid raft disruption

Authors: *M. CACCIOTTOLO[1], A. SAFFARI[2], C. SIOUTAS[2], T. E. MORGAN[1], C. E. FINCH[1]
[1]USC Leonard Davis Sch. of Gerontology, Los Angeles, CA; [2]USC Viterbi Sch. of Engin., Los Angeles, CA

Abstract: Traffic related air pollutants (TRAP) increase the risk of accelerated cognitive decline and Alzheimer Disease (AD). A possible mechanism is the increased levels of brain Aβ, shown in several rodent models. We recently reported that exposure to urban traffic-derived nano-sized

particulate matter (nPM, <0.2 um; PM0.2u) increased Aβ levels in a mouse model of AD, and induced oxidative stress in primary glia culture and in hippocampal slices. Here we investigate the molecular mechanism of nPM induced pro-amyloidogenic APP processing, which we hypothesize are mediated by oxidative stress and lipid raft disruption. Mouse neuroblastoma cells expressing human APP harboring Swedish mutated APP (N2a-APP/swe) were exposed to 1-5-10 μg/ml nPM for 24h. Exposure resulted in dose-dependent increases of Aβ38, -40, and 42 peptides, with increased oxidative stress (NO production, Mitosox, 3-nitrotyrosine -3NT and 4-Hydroxynonenal -4HNE) and altered sub-localization of lipid raft and APP. Treatment with n-acetyl-cysteine (NAC) reduced both oxidative stress and Aβ induced by nPM. We propose that nPM-induced oxidative stress mediates the pro-amyloidogenic processing of APP, with a dose-dependent hierarch. While lower nPM induces rapid NO increase, its increase is below the threshold of oxidative stress required for increased Aβ production.

Disclosures: M. Cacciottolo: None. **A. Saffari:** None. **C. Sioutas:** None. **T.E. Morgan:** None. **C.E. Finch:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.19/X3

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH 5RO1AG048993

NIH 4RO1AG042819

NIH P20GM103442

Title: Comparison of Aβ levels in brains and saliva of Alzheimer’s disease mouse models

Authors: *A. M. FLODEN, G. D. MANOCHA, K. L. PUIG, C. K. COMBS
Biomed. Sci., Univ. of North Dakota, Grand Forks, ND

Abstract: Beta amyloid (Aβ) peptide containing plaque aggregations in the brain are a hallmark of Alzheimer’s Disease (AD). However, Aβ is produced by cell types outside of the brain suggesting that the peptide may serve a broad physiologic purpose. It has been documented that elevated levels of Aβ found can be found in the saliva of AD patients but the source and function of this salivary Aβ is not fully understood. Based upon our prior work documenting expression of amyloid precursor protein (APP) in intestinal epithelium we hypothesized that salivary epithelium might also express APP and be a source of Aβ. To begin testing this idea, we compared C57BL/6 wild type mice to two mouse models of AD, littermate control APP/PS1 mice and the newly characterized APP (NL-G-F) mouse line. At 12-15 months of age, salivary

secretion was stimulated from all three lines and compared to, salivary gland, serum, and brain Aβ levels to determine whether a specific relationship existed between saliva, blood, and brain in either line. As expected, both APP/PS1 and APP (NL-G-F) lines demonstrated robust brain increases in Aβ 1-40/42 with lower levels of Aβ 1-40 in APP (NL-G-F). Both APP/PS1 and APP (NL-G-F) mice also showed an increase in Aβ 1-42 levels in serum with higher Aβ 1-40 levels in APP/PS1 compared to APP (NL-G-F) mice. On the other hand, significantly increased levels of Aβ 1-42 were quantified from saliva of only APP (NL-G-F) mice. These data support the idea that a ratiometric relationship exists between saliva, blood, and brain Aβ levels and indicate that AD mouse models may be useful for exploring the diagnostic potential of this relationship as well as the source and function of salivary Aβ.

Disclosures: A.M. Floden: None. **G.D. Manocha:** None. **K.L. Puig:** None. **C.K. Combs:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.20/X4

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant 6930217

Glenn Foundation

Title: Morphological characterization of glia reveals circuit specific propagation of amyloid beta pathology in Alzheimer's disease

Authors: *F. ABDURROB, R. CANTER, N. DEDIC, K. CHUNG, L.-H. TSAI
Picower Inst. for Learning and Memory, Dept. of Brain and Cognitive Sci., MIT, Cambridge, MA

Abstract: Glia play a crucial role in maintaining homeostasis in the brain and supporting the functioning of neurons. A complex network of inflammatory and neurodegenerative responses, coupled with amyloid beta (Aβ) deposits, tau tangles, and oxidative stress characterize Alzheimer’s disease (AD). Activation and proliferation of microglia and astrocytes are important in the inflammatory response, which ultimately affects disease progression. Consequently, the synergistic changes that occur during gliosis have become an intense topic of investigation, but remain poorly defined. This study utilizes the 5xFAD AD mouse model, as well as healthy age-matched control animals, to spatiotemporally characterize amyloid spread and glial cell response in the brain as aging occurs. Cutting-edge CLARITY and SWITCH labeling techniques are used to visualize the cellular landscape of the diseased brain and create a spatially unbiased map of

endogenous Aβ accumulation. We observe a network-specific spread of amyloid pathology along the Papez long-term memory circuit as a region of vulnerability to early amyloid aggregation and results in physiological impairment. The 4D nature of these data also allow for the visualization of affliction in the fornix, the long-range white matter tract which connects the major nodes of the Papez circuit: the hippocampus, subiculum, and mammillary body. Morphological and proliferative changes in microglia and astrocytes are found in nodes that harbor increased plaque load. The interaction of these immune cells with the vasculature is also examined to explore potential impacts on amyloid clearance. Ultimately, a clearer picture of glial changes during pathology progression along vulnerable neurocircuits will allow for a better understanding of the dynamic pathophysiology that underpins neurodegeneration.
Funding Sources: NIH (6930217) and the Glenn Foundation.

Disclosures: F. Abdurrob: None. **R. Canter:** None. **N. Dedic:** None. **K. Chung:** None. **L. Tsai:** None.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.21/DP05/X5 (Dynamic Poster)

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Identification of measurable phenotypes relevant to Alzheimer’s disease using human iPSC-derived neurons

Authors: K. MANGAN[1], K. KIM[1], L. HARMS[1], C. KANNEMEIER[1], B. M. BADER[3], K. JÜGELT[3], O. SCHRÖDER[3], *B. FREITAS[2], C. B. CARLSON[1], E. JONES[1]
[1]Cell. Dynamics Intl., Madison, WI; [2]Cell. Dynamics Intl., San Diego, CA; [3]NeuroProof GmbH, Rostock, Germany

Abstract: Alzheimer’s disease (AD) is a progressive neurodegenerative disease that results in gradual memory loss and impairment in the ability to learn or carry out daily tasks. The development of therapies for AD has been hindered by limited availability of relevant cell models for basic research and drug discovery. Using induced pluripotent stem cell (iPSC) technology, we have created an unlimited source of human neurons available for studying the mechanisms of AD progression and to streamline the identification of novel drug treatments for this disease. A hallmark of AD pathology is the development of plaques in the brain that contain toxic beta amyloid peptides (Abeta). Therefore, a key focus of AD research is to discern the specific contributions of Abeta to the disease. We have taken two strategies to generate an iPSC-based “disease-in-a-dish” approach for modeling AD *in vitro*. The first is based on genome engineering of an apparently healthy normal iPSC line to introduce mutations in the gene coding for amyloid precursor protein (APP) and then create human neurons from genetically distinct

samples. We rigorously tested the cell by high content imaging, PCR arrays, biomarker production, and multi-electrode array (MEA). Our data were in general agreement with results observed in other model systems for A673V (known to influence AD progression) and A673T (protective from the disease). Uniquely presented, however, functional assessment on MEA with multi-parametric analysis revealed the APP A673V mutant had a significantly different phenotype than A673T or the isogenic WT control. Secondly, we have examined the effects of exogenous exposure to Abeta peptides. Addition of oligomeric Abeta(1-42) to GABAergic and glutamatergic neurons results in cytotoxicity as read out by ATP and LDH assays. Next, synchronous cultures of excitatory glutamatergic neurons - which can be analyzed on MEA to quantify bursting patterns - display a dose-dependent decrease in network bursting prior to decay in firing rates and subsequent to cell death. Detailed evaluation of the burst structure and action potential morphology will be presented. Importantly, these alterations were not observed in control experiments with Abeta(1-40). Our studies demonstrate the utility iPSC technology to create readily accessible human cell models for AD that recapitulate some of the functional neuronal phenotypes that are associated with this complex disease. Ultimately, the promise is that such gene-associated or in vitro disease models can be used to screen for compounds that rescue these phenotypes and significantly reduce the time and cost to develop new AD therapies and improve patient outcomes.

Disclosures: **K. Mangan:** A. Employment/Salary (full or part-time):; Cellular Dynamics International. **K. Kim:** A. Employment/Salary (full or part-time):; Cellular Dynamics International. **L. Harms:** A. Employment/Salary (full or part-time):; Cellular Dynamics International. **C. Kannemeier:** A. Employment/Salary (full or part-time):; Cellular Dynamics International. **B.M. Bader:** A. Employment/Salary (full or part-time):; NeuroProof GmbH. **K. Jügelt:** A. Employment/Salary (full or part-time):; NeuroProof GmbH. **O. Schröder:** A. Employment/Salary (full or part-time):; NeuroProof GmbH. **B. Freitas:** A. Employment/Salary (full or part-time):; Cellular Dynamics International. **C.B. Carlson:** A. Employment/Salary (full or part-time):; Cellular Dynamics International. **E. Jones:** A. Employment/Salary (full or part-time):; Cellular Dynamics International.

Poster

387. Amyloid-Beta Biochemistry and Toxicity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 387.22/X6

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R01AG051521

R21AG05020

1R01AG053982-01

Title: Traffic-related air pollution (TRAP) mediates amyloidogenesis through TNF signaling

Authors: *A. HAGHANI[1], M. CACCIOTTOLO[1], N. SAFI[1], K. R. DOTY[2], F. SHIRMOHAMMADI[3], B. XIN[4], R. ROHS[4], T. C. TOWN[2], C. SIOUTAS[3], T. E. MORGAN[1], C. E. FINCH[1]
[1]Leonard Davis Sch. of Gerontology, [2]Zilkha Neurogenetic Inst., [3]Viterbi Sch. of Engin., [4]Mol. and Computat. Biol. Program, USC, Los Angeles, CA

Abstract: Traffic-related air pollution (TRAP) accelerates cognitive aging, exacerbates white matter loss, and increases the risk for dementia. In prior studies we showed that TRAP-related nano sized particles (nPM) induces inflammatory responses and amyloidogenesis. Current studies investigate inflammatory signals that are enriched in the cortex transcriptome of the male and female APOE3 or APOE4 transgenic mice exposed to TRAP-related nano sized particles (nPM). This RNA-seq analysis revealed that nPM targets the acute phase response, including TNF pathways, and induces amyloidogenesis pathway at different levels. Based on these results, we hypothesized that TNFα may mediate nPM-induced amyloidogenesis. Accordingly, an in vitro experiment investigated the TNFα signaling pathway in transgenic N2a/APPswe cell line. nPM increased TNFα and APP mRNA in N2a/APPswe cell line, with high correlation. Silencing of TNFR1 by siRNA in N2a/APPswe cells reduced TNFα mRNA levels and ameliorated the upregulation of APP gene during nPM treatment. Next, we screened the TNFR1 downstream transcriptional factors (TFs) that might connect this TNF to APP. Among the analyzed TFs, ZFP, RELA, DAXX, RBCK1 and STAT1 mRNA levels significantly changed following TNFR1 silencing. Further structural bioinformatics analysis and in vitro experiments will document whether any of these TFs have a binding site on APP promotor that can affect the expression of this protein during nPM toxicity. In conclusion, the TNF pathway may be a good target against TRAP-induced amyloidogenesis. Future evaluation of these identified targets in a mouse model exposed to TRAP particles will help us to understand the underlying association between air pollution and Alzheimer disease neuropathology.

Disclosures: A. Haghani: None. **M. Cacciottolo:** None. **N. Safi:** None. **K.R. Doty:** None. **F. Shirmohammadi:** None. **B. Xin:** None. **R. Rohs:** None. **T.C. Town:** None. **C. Sioutas:** None. **T.E. Morgan:** None. **C.E. Finch:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.01/X7

Topic: C.04. Movement Disorders

Title: Early sphingolipid signatures of mutant huntingtin expression in neuronal cell

Authors: *A. M. TRZECIECKA, M. PIQUERAS, S. K. BHATTACHARYA
Ophthalmology, Univ. of Miami, Miami, FL

Abstract: Dysregulation of sphingolipid metabolism has been reported in many neurodegenerative disorders. Yet much remains to be understood about alterations of sphingolipid metabolites in Huntington's disease. Our goal in this study was to provide a picture of the early and thus preferentially primary consequences of mutant huntingtin (htt) expression on neuronal cell sphingolipidome.
We used stable doxycycline-inducible neuronal (PC12) cell lines that express htt exon 1 with 23 (control) or 74 (mutant htt) CAG repeats length. Post-mitotic cells (6 day differentiation with 100 ng/mL NGF under low-serum conditions) were induced with 1 µg/mL doxycycline for 8 h. Samples were spiked with internal standards and lipids extracted using Bligh&Dyer method with alkaline hydrolysis. Untargeted profiling analysis was carried out using reverse phase liquid chromatography coupled to high-resolution mass spectrometer (Q Exactive). Subsequently, peak extraction, identification and alignment were performed using Lipid Search 4.1 software. Quantification is based on the ratio to respective internal standard and normalized to total DNA content.
Based on MS/MS spectra we identified a total of 831 species from 8 sphingolipid subclasses, of which 383 (46%): 132 ceramide (Cer), 36 hexosyl-ceramide (CerG1), 29 dihexosyl-ceramide (CerG2), 96 ceramide 1-phosphate (CerP), 3 lysosphingomyelin(LSM) , 67 sphingomyelin (SM), 15 sphingoid base (So) and 5 sphingoid base 1-phosphate (SoP) species changed significantly upon mutant htt expression (t-test $p<0.05$). We found increase in total concentrations of Cer, CerG1, CerG2, LSM, SM, SoP, and decrease in CerP and So (t-test $p<0.05$). Interestingly, contrary to all other subclasses that followed up- or down-regulation pattern, we observe two distinct populations of SM differentially regulated upon mutant htt expression - increasing SM (45%) and decreasing SM (55%). A closer look at these two reveal structural differences in backbone sphingoid base composition and saturation of fatty acid chains.
In conclusion, profound alterations in sphingolipid profile occur as early events upon mutant htt expression in neuronal cell. However, further studies are needed to elucidate functional significance of these findings.

Disclosures: A.M. Trzeciecka: None. **M. Piqueras:** None. **S.K. Bhattacharya:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.02/X8

Topic: C.04. Movement Disorders

Support: CHDI

Title: Developing and testing of small molecules that specifically degrade HDAC4 protein

Authors: H. A. WILKINSON[1], G. MCALLISTER[3], O. LAZARI[3], N. MACABUAG[3], L. URBONAS[3], M. EZNARRIAGA[3], R. JARVIS[4], P. BRECCIA[4], R. VAN DE BOSPOORT[5], *D. MACDONALD[2], E. DOHERTY[1], T. VOGT[1], I. MUNOZ-SANJUAN[1], C. DOMINGUEZ[1]
[1]CHDI Management, Inc., Princeton, NJ; [2]CHDI Management, Inc., Los Angeles, CA; [3]Biol., [4]Charles River, Cambridge, United Kingdom; [5]Charles River, Leiden, Netherlands

Abstract: Huntington's disease (HD) is a lethal autosomal dominant neurodegenerative disorder resulting from a polyglutamine-encoding CAG expansion in the Huntingtin (*HTT*) gene. HDAC4 co-localizes with mutant HTT (mHtt) cytoplasmic inclusions in two HD mouse models (the R6/2 fragment model and the *Hdh*Q150 full length knock-in model) and genetic reduction of HDAC4 delayed mHTT aggregation in CNS tissue. HDAC4 genetic reduction in the R6/2 HD mouse model ameliorated motor and CNS neurophysiological deficits and improved survival.
This led us to develop several small molecule deacetylase inhibitors suitable for pre-clinical testing of the hypothesis that inhibition of HDAC4 activity would alter aggregation kinetics and phenotypic endpoints in HD animal models. In addition to direct HDAC4 inhibition, we have conjugated an exemplar of our lead series to an E3 Ubiquitin ligase ligand in order to generate a proteolysis targeting chimera (PROTAC) tool which can be used to reduce total HDAC4 protein levels.
We validated that the new chimeric molecules were direct HDAC Class IIa (HDAC4, 5, 7 & 9) deacetylase inhibitors in recombinant biochemical assays and retained the ability to inhibit Class IIa HDACs in a variety of mammalian cell lines. Surprisingly, these PROTAC molecules acted as selective and potent degraders of HDAC4, with DC50 values in the low nM range in a variety of cell types when tested by western blot and MSD assay.
Testing the molecules in human ES cell derived and primary neuronal cells showed potent knockdown of HDAC4 levels in agreement with the data from non-neuronal cell types. We are now working towards proof of principle testing of these molecules *in vivo* in rodent models.
This specific protein degradation approach will ultimately be compared with the effects of direct deacetylase catalytic site inhibitors to see if the PROTAC approach more closely replicates the prior results showing genetic Hdac4 hemizygous knock-out ameliorates the phenotype of a transgenic HD mouse model.

Disclosures: **H.A. Wilkinson:** A. Employment/Salary (full or part-time):; CHDI. **G. McAllister:** A. Employment/Salary (full or part-time):; Charles River. **O. Lazari:** A. Employment/Salary (full or part-time):; Charles River. **N. Macabuag:** A. Employment/Salary (full or part-time):; Charles River. **L. Urbonas:** A. Employment/Salary (full or part-time):; Charles River. **M. Eznarriaga:** A. Employment/Salary (full or part-time):; Charles River. **R. Jarvis:** A. Employment/Salary (full or part-time):; Charles River. **P. Breccia:** A. Employment/Salary (full or part-time):; Charles River. **R. van de Bospoort:** A. Employment/Salary (full or part-time):; Charles River. **D. Macdonald:** A. Employment/Salary (full or part-time):; CHDI. **E. Doherty:** A. Employment/Salary (full or part-time):; CHDI. **T. Vogt:** A. Employment/Salary (full or part-time):; CHDI. **I. Munoz-Sanjuan:** A. Employment/Salary (full or part-time):; CHDI. **C. Dominguez:** A. Employment/Salary (full or part-time):; CHDI.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.03/X9

Topic: C.04. Movement Disorders

Support: Spanish Ministerio de Economía y Competitividad (SAF2014-57160-R)

Fundació la Marató de TV3 (514/C/2014)

ISCIII-Subdirección General de Evaluación and European Regional Development Fund (ERDF): RETICS (RD12/0019/0002)

ISCIII-Subdirección General de Evaluación and European Regional Development Fund (ERDF): CIBERNED (2013/08-4)]

European Union's Horizon 2020 research and innovation programme (Grant No 713140)

Title: Functional connectivity and dynamics in Huntington's disease striatal cultures revealed through large-scale calcium imaging

Authors: S. FERNÁNDEZ-GARCÍA[1,3,4], *M. MASANA[1,3,4], J. G. ORLANDI[1,3,4,2,5], G. GARCÍA-DÍAZ BARRIGA[1,3,4], J. SORIANO[2,6], J. ALBERCH[1,3,4]
[1]Dept. of Biomed. Sci., [2]Física de la Matèria Condensada, Univ. of Barcelona, Barcelona, Spain; [3]Inst. d'Investigacions Biomèdiques August Pi i Sunyer, Barcelona, Spain; [4]Ctr. de Investigación Biomédica en Red sobre Enfermedades Neurodegenerativas, Madrid, Spain; [5]Dept. of Physics and Astronomy, Univ. of Calgary, Calgary, AB, Canada; [6]Inst. of Complex Systems (UBICS), Barcelona, Spain

Abstract: Striatum, the main hub of the basal ganglia circuitry, is the most affected region in Huntington’s disease (HD). In HD, mutant-huntingtin (mHtt) causes an excitatory-inhibitory imbalance of the basal ganglia output pathways and induces motor symptoms. Although alterations of striatal Medium-sized Spiny Neurons (MSN) occur at early stages of the disease, little is known about how this translates into functional changes in the network dynamics. Using high-speed, high-resolution calcium imaging, we have recorded simultaneously hundreds of cells from striatal primary cultures in WT and the R6/1 mouse model of HD and characterized the spontaneous activity patterns of every cell as well as their collective activity. Although most of the striatum is composed by inhibitory neurons, we showed that isolated striatal cultures are functionally active, without the need of excitatory inputs. The percentage of active population is ~10% whilst in cortex ~50% of the population showed spontaneous active in basal conditions. At the single cell level, we identified three populations based on their fluorescence activity traces in

both WT and R6/1. Type 1 (fast increase/exponential decay) characteristic of neurons; Type 2 (slow calcium transients) characteristic of astrocytic calcium waves; and Type 3 (non-detectable fluorescence changes). Heterogeneity in cell populations was confirmed by immunocytochemistry against neuronal and glial (MAP2/GFAP) markers. At the population level, both WT and R6/1 include a subset of neurons that display highly coherent activity, indicating the presence of a functional network. Bursting properties of this group are similar between genotypes. Blockade of $GABA_A$ receptors by bicuculline increased number of active neurons while decreased astrocytic signal. Also, the previously reported silent population became active, showing a characteristic neuronal activity profile. The large silent population was inhibited in basal conditions, but the underlying network is revealed through system-wide disinhibition. Indeed, bicuculline boosts coherent activity throughout the culture and increases burst duration and amplitude, but not frequency, in both genotypes. These results indicate that striatal network dysfunction in HD may not arise from local disinhibition but probably from aberrant afferent activity. Therefore, further analysis is required to characterize the contribution of different neurotransmitter systems to the striatal network dynamics in healthy and HD cultures. Understanding functional network alterations mediated by mHtt is fundamental to decipher initial key mechanisms to finally target early symptoms in HD.

Disclosures: S. Fernández-García: None. **M. Masana:** None. **J.G. Orlandi:** None. **G. García-Díaz Barriga:** None. **J. Soriano:** None. **J. Alberch:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.04/X10

Topic: C.04. Movement Disorders

Support: University of Texas, Medical School at Houston, the Department of Neurobiology and Anatomy start up funds

CPRIT Core Facility Support Award RP120092 Proteomic and Metabolomic Core Facility

NCI/ 2P30CA125123-09 Shared Resources Metabolomics core

Funds from Dan L. Duncan Cancer Center (DLDCC)

Title: Inhibiting sphingosine kinase 2 mitigates mutant huntingtin-induced neurodegeneration

Authors: *N. UZOR[1], J. MORUNO MANCHON[1], M. P. BLASCO[1], S. MANNURU[1], N. PUTLURI[2], E. E. FURR-STIMMING[1], A. S. TSVETKOV[1]
[1]McGovern Med. Sch., Houston, TX; [2]Baylor Col. of Med., Houston, TX

Abstract: Huntington disease (HD) is the most common inherited neurodegenerative disorder. It has no cure. The protein huntingtin causes HD, and mutations to it confer toxic functions to the protein that lead to neurodegeneration. Thus, identifying modifiers of mutant huntingtin-mediated neurotoxicity might be a therapeutic strategy for HD. Sphingosine kinases 1 (SK1) and 2 (SK2) synthesize sphingosine-1-phosphate (S1P), a bioactive lipid messenger critically involved in many vital cellular processes, such as cell survival. In the nucleus, SK2 binds to and inhibits histone deacetylases 1 and 2 (HDAC1/2). Inhibiting both HDACs has been suggested as a potential therapy in HD. Here, we found that SK2 is nuclear in primary neurons and, unexpectedly, overexpressed SK2 is neurotoxic in a dose-dependent manner. SK2 promotes DNA double-strand breaks in cultured primary neurons. We also found that SK2 is hyperphosphorylated in the brain samples from a model of HD, the BACHD mice. These data suggest that the SK2 pathway may be a part of a pathogenic pathway in HD. ABC294640, an inhibitor of SK2, reduces DNA damage in neurons and increases survival in two neuron models of HD. Our results identify a novel regulator of mutant huntingtin-mediated neurotoxicity and provide a new target for developing therapies for HD.

Disclosures: N. Uzor: None. **J. Moruno Manchon:** None. **M.P. Blasco:** None. **S. Mannuru:** None. **N. Putluri:** None. **E.E. Furr-Stimming:** None. **A.S. Tsvetkov:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.05/X11

Topic: C.04. Movement Disorders

Support: CHDI

Title: Up-regulation of EAAT2/GLT1 normalizes multiple components of striatal glutamate transmission in the Q175 mouse model of Huntington’s disease

Authors: *C. RANGEL BARAJAS, I. CORONEL-MORALES, G. V. REBEC
Psychological and Brain Sciences, Program in Neurosciences, Indiana Univ., Bloomington, IN

Abstract: Dysregulation of glutamate transmission is a key feature of Huntington’s disease (HD), a dominantly inherited condition characterized by progressive striatal degeneration and dysfunctional corticostriatal circuits. The major glutamate transporter, excitatory amino acid transporter 2 (EAAT2) or its mouse homolog (GLT1), is down-regulated in both HD patients and transgenic mouse models of HD. We previously reported that EAAT2 up-regulation by intravascular administration of recombinant adeno-associated virus serotype 9 (AAV9) reverses some key motor deficits as well as corresponding changes in striatal electrophysiological activity for several months in the heterozygous Q175 mouse model. Here, we determined if AAV9-

mediated up-regulation of EAAT2 also normalizes other components of striatal glutamate transmission, such as the glial cystine/glutamate exchanger (xCT) and the NMDA receptor subunits, NR2A and NR2B. By exchanging intracellular glutamate for cystine, xCT works closely with GLT1 to maintain glutamate homeostasis, which is altered in HD. Moreover, NMDA receptors have been implicated in HD neurodegeneration. For example, although synaptic NMDA receptors activate cellular survival pathways, extra-synaptic NMDA receptors activate pathways that promote cell death (Hardingham and Bading, Nat. Review Neurosci., 11:682-696, 2010). Importantly, these extra-synaptic receptors are preferentially comprised of NR2B subunits, which are increased in the striatum of a pre-symptomatic HD mouse model (Milnerwood et al., Neuron, 65:178-190, 2010). AAV9-EAAT2 or vehicle was administered to Q175 mice or wild-type (WT) controls at 6 or 7 months of age and striatal tissue was evaluated subsequently by western blot at 10 and 14 months of age, when Q175 neurological signs begin to appear. We found that just as the down-regulation of EAAT2 in Q175s was reversed at 10 and 14 months of age, AAV9-EAAT2 also reversed the decreased expression of xCT at the same ages. Interestingly, AAV9-EAAT2 differentially affected the NMDA receptor subunits. Expression of the NR2B subunit, which is elevated in vehicle-treated Q175 relative to WT mice, was significantly decreased at both 10 and 14 months. In contrast, NR2A expression, which declines in vehicle-treated HD mice relative to WT, is at least partially elevated by AAV9-EAAT2. Collectively, these data show that restoration of EAAT2 expression in the Q175 model normalizes multiple components of striatal glutamate transmission and thus could serve as a therapeutic strategy for HD.

Disclosures: C. Rangel Barajas: None. **I. Coronel-Morales:** None. **G.V. Rebec:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.06/X12

Topic: C.04. Movement Disorders

Support: Funded by Teva Pharmaceutical Industries Israel

Title: Transcriptomic analysis of the YAC128 HD mouse model shows disease mechanisms are ameliorated by pridopidine

Authors: R. KUSKO[1], M. GEVA[2], J. DREYMANN[2], M. POULADI[3], *J. ROSS[1], Y. CHA[1], R. ESCALANTE[1], B. ZESKIND[1], D. LAIFENFELD[2], A. ORBACH[2], R. LAUFER[2], I. GROSSMAN[2], M. R. HAYDEN[2]
[1]Immuneering Corp., New York, NY; [2]Teva Pharmaceut. Industries Ltd, Netanya, Israel; [3]Natl. Univ. of Singapore, Singapore, Malaysia

Abstract: Huntington Disease (HD) is a neurodegenerative disorder hallmarked by the expression of a mutant form of the huntingtin gene (mHtt). A therapeutic goal for HD treatment involves the restoration of neurobiological pathways disrupted by mHtt. Pridopidine, an investigational HD drug candidate, has been shown to improve motor symptoms in both preclinical models as well as in HD patients. While originally described as a dopaminergic stabilizer, in vitro binding studies show highest affinity of pridopidine to the sigma-1 receptor. We previously reported that pridopidine upregulates BDNF, glucocorticoid receptor (GR), and dopamine 1 receptor (D1R) signaling pathways in WT rat striatum. To expand on these findings and explore molecular changes specific to HD, we repeated this study in a YAC128 HD mouse model whereby mice were treated with pridopidine or vehicle starting at postnatal week 6 and sacrificed after 11.5 months of age. We then performed RNAseq data analysis on the striatum. We identified 1346 differentially expressed genes (DEGs) in vehicle treated YAC128 mice vs. healthy controls, 221 DEGs in YAC128 mice treated with 30 mg/kg of pridopidine vs. vehicle, and 73 DEGs in YAC128 mice treated with 10 mg/kg of pridopidine vs. vehicle (all adj.$p<0.05$). In addition, the previously reported pridopidine-induced upregulation of BDNF, GR, and D1R pathways was confirmed after treatment with either dose (adj.$p<0.05$), and alternatively spliced genes after 30 mg/kg of pridopidine compared to YAC128 vehicle are enriched for the BDNF pathway (adj.$p<0.05$). Further, pathway analysis of DEGs after 30 mg/kg dosage revealed enrichment for biological processes related to synaptic transmission, including upregulation in the key HD-impaired processes - cAMP and calcium signaling (adj.$p<0.05$). Lastly, since mHtt was shown to induce neurotoxicity by promoting M1 microglia that secrete proinflammatory cytokines, enrichment analysis tested for microglial markers. Treatment with 30 mg/kg of pridopidine led to the downregulation of genes associated with M1 activation (adj.$p<0.05$). To summarize, pridopidine induces transcriptional modifications reversing genes in HD-impaired pathways involved in neuronal transmission and protection in the YAC128 striatum.

Disclosures: **R. Kusko:** A. Employment/Salary (full or part-time):; Teva Pharmaceutical Industries Ltd. **M. Geva:** None. **J. Dreymann:** None. **M. Pouladi:** A. Employment/Salary (full or part-time):; Teva Pharmaceutical Industries Ltd. **J. Ross:** A. Employment/Salary (full or part-time):; Teva Pharmaceutical Industries Ltd. **Y. Cha:** A. Employment/Salary (full or part-time):; Teva Pharmaceutical Industries Ltd. **R. Escalante:** A. Employment/Salary (full or part-time):; Teva Pharmaceutical Industries Ltd. **B. Zeskind:** A. Employment/Salary (full or part-time):; Teva Pharmaceutical Industries Ltd. **D. Laifenfeld:** None. **A. Orbach:** None. **R. Laufer:** None. **I. Grossman:** None. **M.R. Hayden:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.07/X13

Topic: C.04. Movement Disorders

Support: CHDI A-9929

Title: Zinc finger protein reduces mHtt expression and attenuates qEEG oscillatory dysfunction in HD mice

Authors: *E. J. DONZIS[1], K. SAFARYAN[2], N. BERBERIAN[1], G. GANESH[1], S. CHOPRA[3], G. KONDAVEETI[3], K. REINA[3], A. M. ESTRADA-SANCHEZ[1], A. M. HUNTER[3], C. CEPEDA[1], M. R. MEHTA[2], A. F. LEUCHTER[3], M. S. LEVINE[1]
[1]IDDRC, Semel Inst. for Neurosci. and Human Behavior, BRI, [2]Departments of: Physics & Astronomy, Neurology, Neurobio., [3]Psychiatry and Biobehavioral Sciences, TMS Clin. and Res. Service, Neuromodulation Div., Univ. of California Los Angeles, Los Angeles, CA

Abstract: Huntington's disease (HD) is an autosomal dominant genetic disorder characterized by cognitive and motor deficits. Currently, there is no cure for HD, only therapies to relieve symptoms. Since HD is progressive and has a generally late-onset, isolating early and reliable markers of the disease is important for targeted therapeutic intervention. Electrophysiological dysfunction long precedes neuronal death and may therefore serve as a useful biomarker. Specifically, EEG oscillatory disturbances are seen in premanifest gene-positive patients and may serve as an early biomarker. Identification of these biomarkers may be useful in testing the efficacy of potential treatment strategies. We used Q175 heterozygotic mice to measure disturbances in oscillatory synchrony in basal ganglia and cortical circuits during disease progression. Using 4 different age groups (8-18 weeks, 19-29 weeks, 30-40 weeks, and 41-51 weeks) spanning presymptomatic through symptomatic stages and recording EEGs/LFPs from the striatum, thalamus, frontal cortex, and occipital cortex, we found age-dependent disruptions in relative delta power in the striatum and thalamus as well as a disruption in the anterior posterior gradient of qEEG power at the delta and alpha frequencies. After identifying these alterations, the striatum was injected with a zinc finger protein (ZFP), thought to reduce mutant huntingtin aggregates (mHtt), in a separate cohort of mice (aged 30-50 weeks). We verified a 30% reduction of mHtt aggregates in the striatum 8 weeks post ZFP injection. The mHtt aggregate reduction was associated with an attenuation of the EEG oscillatory dysfunction seen in the basal ganglia and cortex of HD mice. Overall, these studies suggest that disruptions in oscillatory synchrony may be a useful early biomarker in HD and that ZFP-induced reduction in mHtt aggregates is a promising treatment strategy.

Disclosures: E.J. Donzis: None. **K. Safaryan:** None. **N. Berberian:** None. **G. Ganesh:** None. **S. Chopra:** None. **G. Kondaveeti:** None. **K. Reina:** None. **A.M. Estrada-Sanchez:** None. **A.M. Hunter:** None. **C. Cepeda:** None. **M.R. Mehta:** None. **A.F. Leuchter:** None. **M.S. Levine:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.08/X14

Topic: C.04. Movement Disorders

Support: CHDI A-8462

Title: Striatal GABAergic interneurons show distinct electrophysiological changes during disease progression in the Q175 mouse model of Huntington's disease

Authors: *S. M. HOLLEY, L. GALVAN, T. KAMDJOU, C. CEPEDA, M. S. LEVINE
IDDRC, Semel Inst. for Neurosci. and Human Behavior, BRI, UCLA, Los Angeles, CA

Abstract: Postmortem analysis of patients with Huntington's disease (HD) has shown extensive degeneration of striatal medium-sized spiny neurons (MSNs), the major output projection neurons in the striatum. MSN degeneration has been well characterized in several mouse models of HD, including the Q175 knock-in model that contains the human mutant huntingtin allele (mHtt) with an expanded CAG tract (~175 repeats) within the native mouse huntingtin gene. However, few reports describe how striatal GABAergic interneurons are affected in these mice as the disease progresses. In order to assess how interneurons may contribute to the HD phenotype, we crossed heterozygous Q175 mice with Lhx6-EGFP mice allowing for visualization of fast-spiking interneurons (FSIs) and persistent low-threshold spiking (PLTS) interneurons. Using whole-cell patch clamp electrophysiology, we characterized these two populations of GABAergic interneurons in presymptomatic (2-3 month-old) and symptomatic (12 month-old) mice. In 12 month-old mice we found FSIs exhibited abnormal passive membrane properties (decreased cell capacitance, increased input resistance and decreased membrane time constant), suggestive of degenerative changes in symptomatic mice, while PLTS interneurons appeared spared. However, both types of interneurons displayed increased excitability. The resting membrane potential was more depolarized in Q175 FSIs compared to wild-type at only the symptomatic age and input-output curves showed increased firing in these interneurons. Moreover, PLTS interneurons, that normally display spontaneous firing, exhibited an increase in the frequency of burst firing in symptomatic mice when compared to wild-types. We also showed that Q175 FSIs receive fewer spontaneous excitatory synaptic inputs at the older age, while inhibitory inputs remained unchanged throughout the disease progression. There were no differences in the frequency of spontaneous synaptic input in PLTS interneurons at both ages tested. Based on these observations, we conclude that both FSIs and PLTS interneurons are distinctly affected in this HD model as the disease progresses, which ultimately can lead to an imbalance in inhibitory tone in the HD striatum.

Disclosures: **S.M. Holley:** None. **L. Galvan:** None. **T. Kamdjou:** None. **C. Cepeda:** None. **M.S. Levine:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.09/X15

Topic: C.04. Movement Disorders

Support: SAF-2014-57160R

SAF2015-67474-R;MINECO/FEDER

CIBERNED

Title: Cdk5 contributes to Huntington's disease learning and memory deficits via modulation of brain region-specific substrates

Authors: E. ALVAREZ-PERIEL[1,2,3], M. PUIGDELLIVOL[1,2,3], V. I. BRITO[1,2,3], F. PLATTNER[4], J. A. BIBB[5], J. ALBERCH[1,2,3], *S. GINES-PADROS[1,2,3]
[1]Med. School, Univ. of Barcelona, Barcelona, Spain; [2]IDIBAPS, Barcelona, Spain; [3]CIBERNED, Madrid, Spain; [4]Dept. of Psychiatry, UT Southwestern Med. Ctr., Dallas, TX; [5]Surgery, Univ. of Alabama Birmingham, Birmingham, AL

Abstract: Cognitive deficits are a major hallmark of Huntington´s Disease (HD) with a great impact on the quality of patient's life. Gaining a better understanding of the molecular mechanisms underlying learning and memory impairments in HD is therefore, of critical importance. Cdk5 is a proline-directed Ser/Thr kinase involved in the regulation of synaptic plasticity and memory processes that has been associated with several neurodegenerative disorders. In this study, we aim to investigate the role of Cdk5 in learning and memory impairments in HD using a novel animal model that expresses mutant huntingtin (mHtt) and has genetically reduced Cdk5 levels. Genetic reduction of Cdk5 in mHtt knock-in mice attenuated both corticostriatal learning deficits as well as hippocampal-dependent memory decline. Moreover, the molecular mechanisms by which Cdk5 counteracts the mHtt-induced learning and memory impairments appeared to be differentially regulated in a brain region-specific manner. While the corticostriatal learning deficits are attenuated through compensatory regulation of NR2B surface levels, the rescue of hippocampal-dependent memory was likely due to restoration of hippocampal dendritic spine density along with an increase in Rac1 activity. This work identifies Cdk5 as a critical contributor to mHtt-induced learning and memory deficits. Furthermore, we show that the Cdk5-downstream targets involved in memory and learning

decline differ depending on the brain region analysed suggesting that distinct Cdk5 effectors could be involved in cognitive impairments in HD.

Disclosures: E. Alvarez-Periel: None. **M. Puigdellivol:** None. **V.I. Brito:** None. **F. Plattner:** None. **J.A. Bibb:** None. **J. Alberch:** None. **S. Gines-Padros:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.10/X16

Topic: C.04. Movement Disorders

Support: Huntington Society of Canada

Memorial University Dean's Innovation Fund

Title: Characterization of the onset and severity of synaptic plasticity deficits in the Q175FDN knock-in mouse model of Huntington's disease

Authors: *J. G. QUIRION, M. P. PARSONS
Biomed. Sci., Mem. Univ. of Newfoundland, St John's, NL, Canada

Abstract: Huntington’s Disease (HD) is a progressive neurodegenerative disease characterized by motor, psychiatric and cognitive disturbances and is caused by a CAG repeat in the first exon of the gene encoding the huntingtin protein. Few studies have examined the cellular mechanisms underlying the cognitive deficits in HD, although many patients report the cognitive symptoms to be the most debilitating. Currently, there is a poor understanding of how the disease-causing mutant huntingtin protein affects cognition at the cellular level. Changes in synaptic plasticity in the hippocampus are thought to underlie cognitive processes such as learning and memory. As such, studying hippocampal synaptic plasticity can foster an understanding of the neural underpinnings of cognitive deficits in HD. This study aimed to examine the progression of synaptic correlates of cognitive decline in HD by studying hippocampal long term potentiation (LTP) at different stages of disease progression in Q175FDN mice, a slowly-progressing knock-in mouse model of HD with clear cognitive deterioration in both heterozygous and homozygous mutation carriers. Golgi staining was also used to examine changes in pyramidal neuron spine density and morphology in order to relate these findings to any observed changes in synaptic excitability and plasticity. Surprisingly, despite the high expression level of mutant huntingtin and the clear cognitive deficit at 9 months of age in this model, heterozygous Q175FDN mice had intact LTP, spine density and spine morphology at three, six, and nine months of age. In stark contrast, homozygote Q175FDN mice showed clear deficits in LTP starting at three months as well as an increase in basal excitability and spine density that were later decreased at nine

months. Thus, the LTP deficit in homozygote mice is unlikely to be explained solely by weak synaptic connectivity under basal conditions. As Q175FDN mice have higher expression levels of mutant huntingtin compared to prior knock-in models, the dramatic difference in synaptic plasticity between heterozygous and homozygous mice suggests a potential protective role for the presence of non-pathogenic huntingtin in the heterozygous mice. Future studies will explore the possibility that long-term reductions in non-pathogenic huntingtin may impact synaptic plasticity. In all, our data suggests that the plasticity deficits in the HD hippocampus are complex and are heavily impacted by the expression level of mutant huntingtin and/or the loss of non-pathogenic huntingtin.

Disclosures: J.G. Quirion: None. **M.P. Parsons:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.11/X17

Topic: C.04. Movement Disorders

Support: Taube Philanthropies

Koret Foundation 12-0160

Jean Perkins Foundation

National Institute of Mental Health

Title: TSPO-PET imaging using ^{18}F-PBR06 is a potential biomarker for monitoring therapeutic Efficacy in Huntington's Disease: preclinical evidence with the small molecule p75^{NTR} ligand LM11A-31

Authors: *D. A. SIMMONS[1], M. L. JAMES[2], N. P. BELICHENKO[1], S. SEMAAN[1], C. CONDON[1], J. C. KUAN[1], A. SHUHENDLER[2], Z. MIAO[2], F. T. CHIN[2], F. M. LONGO[1]
[1]Neurol. and Neurolog. Sci., [2]Dept. of Radiology, Mol. Imaging Program, Stanford Univ., Stanford, CA

Abstract: Huntington's Disease (HD) is a fatal neurodegenerative disorder caused by an expanded CAG repeat in the gene encoding the huntingtin protein. No therapy exists for delaying the onset or slowing the progression of HD. The search for disease-modifying treatments would benefit from non-invasive biomarkers that monitor disease progression and therapeutic efficacy in HD mouse models and human trials. One potential biomarker is examining translocator protein 18kDa (TSPO) by positron emission tomography (PET) imaging to detect microglial activation, a key contributor to HD neurodegeneration. PET imaging with [^{11}C]PK11195, a first

generation TSPO tracer with poor specificity and sub-optimal brain permeability, detected activated microglia in pre-symptomatic HD patients and correlated with disease onset and severity. Second generation TSPO-PET radiotracers (e.g., [^{18}F]PBR06) with improved sensitivity have been developed, but have yet to be evaluated in HD patients. Moreover, whether TSPO-PET can detect microglial activation in HD mouse models or monitor therapeutic efficacy in HD is unknown. Thus, the ability of [^{18}F]PBR06-PET to detect microglial activation and to monitor treatment response of the p75^{NTR} ligand LM11A-31, previously shown to reduce microglial activation in HD mice, was assessed in two HD mouse models. LM11A-31 was orally administered (50 mg/kg, daily 5 days/week) to R6/2 and BACHD mice for 7 weeks and 7 months, respectively. [^{18}F]PBR06-PET/CT images were acquired at a late disease stage for R6/2 mice (11-12 weeks old) and early and mid-symptomatic stages for BACHD mice (5 and 9 months old). [^{18}F]PBR06-PET signal was elevated in the striatum, cortex and hippocampus of vehicle-treated R6/2 mice versus age-matched wild-types and LM11A-31-treated R6/2 mice had significantly reduced [^{18}F]PBR06 accumulation. Notably, [^{18}F]PBR06-PET signal was also significantly increased in mid-, but not early, symptomatic BACHD mice in all brain areas examined and ameliorative effects of LM11A-31 on neuroinflammation were discerned. In both mouse models, [^{18}F]PBR06-PET signal correlated with increased immunostaining for IBA-1, an activated microglia marker, and western immunoblotting for TSPO. These results are the first to show that TSPO-PET imaging can track microglial activation and monitor therapeutic efficacy in HD mice, indicating that it is a potential translatable biomarker for preclinical to clinical HD testing. Since LM11A-31 is currently in phase IIa clinical trials for Alzheimer's disease, having a biomarker capable of detecting therapeutic efficacy, as shown here with TSPO-PET, could expedite its advancement to HD clinical trials.

Disclosures: D.A. Simmons: None. **M.L. James:** None. **N.P. Belichenko:** None. **S. Semaan:** None. **C. Condon:** None. **J.C. Kuan:** None. **A. Shuhendler:** None. **Z. Miao:** None. **F.T. Chin:** None. **F.M. Longo:** Other; Dr. Longo is listed as an inventor on patents relating to a compound in this report, which is assigned to the University of North Carolina, University of California, San Francisco and the Dept. of Vet.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.12/X18

Topic: C.04. Movement Disorders

Support: NIH Grant NS040408

Title: Neuroprotection by NF-κB subunit RelA/P65 in cell culture models of Huntington's disease and spinocerebellar ataxia type-1

Authors: *Y. ZHANG, ***Y. ZHANG**, J. FRANKLIN, S. D'MELLO
Biol. department, Southern Methodist Univ., Dallas, TX

Abstract: We and other labs and previously described that RelA/p65, a subunit of the NF-kB transcription factor, protects cultured neurons against death induced by a variety of apoptosis-inducing stimuli. We have now extended these studies to cell culture models of neurodegenerative diseases. We report that overexpression of p65 is protective in cell culture models of Huntington's disease (HD) and Spinocerebellar ataxia type 1(SCA1) utilizing cortical and cerebellar granule neurons, respectively. We previously described that expression of endogenous p65 is not altered in various paradigms of neuronal apoptosis. We find that expression was not discernibly altered in the striatum of R6/2 mice, a commonly used mouse model of HD. we investigated whether posttranslational modifications including acetylation and phosphorylation sites or protein-protein interactions were involved. Mutation of five different acetylation sites either individually or in combination had no effect on mut-Htt-mediated neuroprotection Similarly, mutation of two separate phosphorylation sites, Ser 467 and Ser 534, had no effect. Co-immunoprecipitation analyses showed that p65 interacted with huntingtin (Htt), but not with SCA-1, a finding that was confirmed through co-localization studies. While interacting between p65 and wild-type Htt was strong, it was lower with mutant-Htt. Further analysis using deletion constructs localized interaction of p65 to the 1 - 75 amino acid region of Htt. Current research is aimed at understanding in more detail the mechanism by which p65 protects against neurotoxicity by mutant-Htt and mutant-SCA1.

Disclosures: Y. Zhang: None. **J. Franklin:** None. **S. D'Mello:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.13/X19

Topic: C.04. Movement Disorders

Support: NIH Grant NS040408

Title: Reduced expression of Foxp1 as a contributing factor in Huntington's disease

Authors: *A. LOUIS SAM TITUS[1], S. R. D'MELLO[2]
[1]Dept. of Mol. and Cell Biol., UNIV OF TEXAS AT DALLAS, Dallas, TX; [2]Biol. Sci., Southern Methodist Univ., Dallas, TX

Abstract: Huntington's disease (HD) is an inherited neurodegenerative disease caused by a polyglutamine expansion in the huntington protein (*htt*). The neuropathological hallmark of HD is the loss of neurons in the striatum and, to a lesser extent, in the cortex. *Foxp1* is a member of

the Forkhead family of transcription factors expressed selectively in the striatum and the cortex. In the brain, three major *Foxp1* isoforms are expressed - isoform-A (~90 kDa), isoform-D (~70 kDa) and isoform-C (~50 kDa). We find that expression of *Foxp1* isoforms A and D is selectively reduced in the striatum and cortex of R6/2 HD mice as well as in the striatum of HD patients. Furthermore, expression of mutant *htt* in neurons results in the downregulation of *Foxp1*. Elevating expression of isoform A or D protects cortical neurons from death caused by the expression of mutant *htt*. On the other hand, knockdown of *Foxp1* promotes death in otherwise healthy neurons. Neuroprotection by *Foxp*1 is likely to be mediated by the transcriptional stimulation of the cell cycle inhibitory protein, $p21^{Waf1/Cip1}$. Consistently, *Foxp1* activates transcription of the $p21^{Waf1/Cip1}$ gene promoter and overexpression of *Foxp1* in neurons results in the elevation of p21 expression. Moreover, knocking down of $p21^{Waf1/Cip1}$ blocks the ability of *Foxp1* to protect neurons from mut-Htt-induced neurotoxicity. We propose that the selective vulnerability of neurons of the striatum and cortex in HD is related to the loss of expression of *Foxp1*, a protein that is highly expressed in these neurons and required for their survival.

Disclosures: A. Louis Sam Titus: None. **S.R. D'Mello:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.14/X20

Topic: C.04. Movement Disorders

Support: Mass based Biogen Idec.and the sastry foundation

Title: Striatal and cortical changes in huntington disease

Authors: *W. SWEIDAN[1], N. SERAJI BOZORGZAD[1], F. BAO[2], K. ARRAGUNTLA[1], S. LITCHMAN[1], S. RAZMJOU[1]
[1]Wayne State Univ., Detroit, MI; [2]Univ. Hlth. Ctr., Detroit, MI

Abstract: OBJECTIVES: To assess cortical microstructural changes in Huntington's disease (HD) patients and its correlation with cortical thickness and striatal volumes**,** using surface based laminar analysis of diffusion abnormalities. **METHODS:** 6 symptomatic HD patients (CAG repeats: median=45.5; IQR= 4; 55± 10.72 years) and 6 healthy controls (HC, 33±10.7years) underwent Magnetic Resonance Imaging to assess cortical lobes thickness and striatal volumes, Magnetic Resonance Spectroscopy to examine neuronal integrity in deep white matter structure, centrum semiovale (8x8 voxel), and diffusion tensor imaging (DTI) to assess white matter (WM) and grey matter (GM) diffusivity. **RESULTS:** As shown previously, HD patients presented evidence of neuronal loss compared to HC, reflected by reduced average cortical thickness

(frontal: 2.4±0.07mm v. 2.5±0.1; parietal: 2.1±0.2mm v. 2.4±0.1; occipital: 1.9±0.2mm v. 2.1±0.08 lobes), reduced subcortical volumes (caudate: 4.0±1.3mm^3 v. 7.7±1.2mm^3; putamen: 6.0±2.8 mm^3 v. 11.8±0.6 mm^3; pallidum: 1.8±0.6 mm^3 v. 3.4±0.3 mm^3), and reduced N-acetylaspartate to creatinine ratio (1.6±0.2 v. 2.0±0.1). Disruption of tissue architecture was also evidenced by higher radial diffusivity of whole brain WM in HD compared to HC (p=0.03), consistent with previous findings. Moreover, we were able to demonstrate loss of cortical tissue integrity using surface based laminar analysis of GM DTI parameters. A marked increase in axial (AD), radial (RD) and mean diffusivity (MD) was reported in the frontal (p=0.02, 0.01, and 0.02, respectively), temporal (p=0.03, 0.03, and 0.03, respectively), parietal (p=0.03, 0.02 and 0.04, respectively) and occipital lobes (p=0.03, 0.02, and 0.03, respectively) in the HD v. HC. Furthermore, DTI measures were significantly correlated with cortical thickness in the parietal (AD: r=-0.99, p=0.002; RD: r=-0.92, p=0.005; MD: r=-0.99, p=0.002), temporal (MD: r=-0.9, p=0.04; AD: r=-0.9, p=0.04), and occipital lobes (AD: r=-0.95, p=0.01; RD: r=-0.95, p=0.01; MD: r=-0.99, p=0.002). **CONCLUSION:** HD group showed higher cortical MD, AD, and RD, possibly reflecting microstructural changes such as dendritic and synaptic remodeling, and cytoskeletal alterations at the level of cortical neurons prior to degeneration. Diffusivity changes were evident in temporal lobe prior to its thinning, suggesting potential role as an earlier marker to track disease progress.

Disclosures: W. Sweidan: None. **N. Seraji Bozorgzad:** None. **F. Bao:** None. **K. arraguntla:** None. **S. litchman:** None. **S. razmjou:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.15/X21

Topic: C.04. Movement Disorders

Support: NIH Grant U44 NS090616

Azevan Pharmaceuticals, Inc

Title: Irritability in Huntington's Disease: A phase II exploratory clinical trial with a novel vasopressin 1a antagonist, SRX246

Authors: *S. M. HERSCH[1], ***S. M. HERSCH**[1], S.-F. LU[2,3], K. E. ANDERSON[4], M. J. BROWNSTEIN[2], N. G. SIMON[2,3]
[1]Dept Neurol., Massachusetts Gen. Hosp. Dept. of Neurol., Charlestown, MA; [2]Azevan Pharmaeuticals, Inc., Bethlehem, PA; [3]Biol. Sci., Lehigh Univ., Bethlehem, PA; [4]Psychiatry and Neurol., Georgetown Univ. Med. Ctr., Washington, DC, DC

Abstract: Psychiatric symptoms, including irritability and aggression, are common in HD patients. These are among the most distressing aspects of the disease, adversely impact daily life, and often result in institutionalization. Despite their frequent occurrence and severe consequences, these symptoms have received little attention. Effective treatments are lacking and well-validated scales for measuring changes in these symptoms are not available.
The current Phase II clinical trial in HD patients (n=108), Safety and Tolerability of SRX246 In Irritable/Aggressive Subjects with Huntington's Disease (NCT02507284), is designed to rigorously evaluate the tolerability of a new drug, SRX246, for the treatment of irritability and aggression; provide additional safety data; and explore various rating scales for the assessment of changes in these symptoms. The objective is to obtain critical data that can be used to plan future Phase IIb or III clinical trials. STAIR is a 3 arm, multicenter (22 NeuroNEXT Network sites), randomized, placebo-controlled, double-blind, 12-week dose escalation study. Following eligibility determination, subjects are randomized to receive placebo, or escalate from 80 mg (two weeks) to 120 mg, up to a maximum dose 160 mg twice daily of SRX246 for 12 weeks. Each subject has a study partner to assist with visits, taking study medication, and providing feedback about the subject's mood and behavior. As of May 1, 51 subjects were randomized, 23 have completed the protocol, and 16 additional individuals have consented. Compliance has been very good and while we are blinded, the AE profile and tolerability are consistent with other trials that show good safety and tolerability.
The test compound, SRX246, is a first-in-class vasopressin 1a receptor antagonist. It is orally bioavailable, exhibits high affinity and selectivity for its target, has a strong safety profile in healthy volunteers, and excellent pharmacokinetics. Preclinical pharmacology and an experimental medicine fMRI study in healthy volunteers showed that SRX246 has CNS effects after oral administration and that it modulates brain circuits involved in responses to stimuli that elicit aggression/fear. In a recently completed Phase II Exploratory trial for the treatment of Intermittent Explosive Disorder, SRX246 was well tolerated, no serious adverse events were reported, and exploratory analyses revealed statistically significant differences favoring SRX246 in key outcome measures of clinical benefit. These findings strongly suggested that SRX246 might have a beneficial effect on the irritability and aggression seen in a sizable proportion of HD patients.

Disclosures: S.M. Hersch: None. **S. Lu:** A. Employment/Salary (full or part-time):; Azevan Pharmaceuticals, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Azevan Pharmaceuticals, Inc. **K.E. Anderson:** F. Consulting Fees (e.g., advisory boards); Azevan Pharmaceuticals, Inc. **M.J. Brownstein:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NIH grant 1 U44 NS090616. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Azevan Pharmaceuticals, Inc. **N.G. Simon:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Azevan Pharmaceuticals, Inc..

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.16/X22

Topic: C.04. Movement Disorders

Title: Identifying functional regulatory units in Huntington's Disease Mouse Models using a population based evolutionary algorithm

Authors: *S. ALLAM, T. H. RUMBELL, T. TRONG, A. PONZI, J. R. KOZLOSKI
Computat. Biol. Ctr., IBM Res., Yorktown Heights, NY

Abstract: Symptomatic R6/2 mouse model and Q175 exhibit numerous electrophysiological variations from the wild type. Passive and active membrane properties such as (membrane voltage, rheobase, average number of spikes per stimulus, spike onset latency) of MSNs are altered and affect the integrative properties of the basal ganglia circuitry. We used a population based evolutionary algorithm to reproduce the range of biophysical features of wild type and Huntington's disease (HD) mouse model MSNs in a computational MSN model built using IBM's Neural Tissue Simulator. Principal component analysis was performed on those models to identify the key functional regulatory units that transform the wild type membrane properties to the HD phenotype. We discuss how our approach for finding low dimensional descriptions of disease phenotypic conversion in neuronal models may allow target identification for pharmacological intervention to alleviate HD symptoms in these neurons.

Disclosures: S. Allam: None. **T.H. Rumbell:** None. **T. Trong:** None. **A. Ponzi:** None. **J.R. Kozloski:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.17/X23

Topic: C.04. Movement Disorders

Support: Funded by CHDI Foundation

Title: Results of a phenotypic HTS to identify small molecule modulators of mutant HTT levels in hES cells as HD therapies

Authors: O. LAZARI[1], A. MUKONOWESHURO[1], B. NANCOLAS[1], C. TURNBULL[1], I. GOWERS[1], M. IOVINO[1], C. PAULE[1], R. JARVIS[1], P. BRECCIA[1], T. LADDUWAHETTY[1], G. MCALLISTER[1], E. DOHERTY[2], *J. A. BARD[2], D. MACDONALD[2], A. HOWARD[2], C. DOMINGUEZ[2], I. MUNOZ-SANJUAN[2]
[1]Charles River, Cambridge, United Kingdom; [2]CHDI Management, Inc., Princeton, NJ

Abstract: Lowering of the pathogenic mutant huntingtin (mHTT) protein in Huntington's disease (HD) patients is one of the leading approaches to ameliorate the fatal neurodegeneration caused by the poly-CAG expansion in the *HTT* gene.
Although HTT lowering therapies based on siRNA/ASOs/ZFPs are in development, they all have challenges in terms of invasive dosing routes and schedules, and relatively poor distribution to overcome. Therefore, identification of brain penetrant small molecules, with convenient oral dosing and systemic distribution, would potentially be advantageous over these novel biological agents.
Previous efforts to identify small molecule HTT lowering agents have generally used highly expanded HTT polyQ repeats, non-specific readouts such as cytotoxicity or aggregation, or used the *HTT* gene in a non-native genomic context. These efforts have not delivered clinically relevant candidates.
In an attempt to identify novel agents reducing disease and physiologically relevant mHTT levels, we have developed a phenotypic assay suitable for HTS of large compound collections. HD human ES (hES) cells, with a clinically-relevant CAG expansion [Q48] HTT allele under the control of the endogenous promoter, are plated in a 384-well plate and challenged with small molecules for 48 h. Cells are then lysed and mHTT and total HTT are detected by sensitive antibodies to epitopes in the N-terminal of the protein in a HTRF format. Toxicity is monitored by ATP determination as a surrogate for cell number. Assay statistics (robust Z' >0.4; S:B >4) supported using the assay in an HTS format.
As these assays are able to measure changes in the downstream HTT protein levels, they are not biased to identify molecules which only affect a hypothetical endpoint of HTT toxicity (e.g., aggregation) or a single mode of action (e.g., via changes in transcription, translation, or clearance).
We used this assay to screen a diverse compound library of ~120K compounds from the CHDI collection. Hit matter was reliably detected and showed good translation in an IC50 format, where compounds mediating effects via significant toxicity were removed from the screening cascade.
A screening cascade for hits with a window over toxicity to potentially define different mechanisms of action was used, culminating in the use of HD hES-derived neuronal cells from the primary screening hES line to measure whether the effects of hits are maintained in a neuronal phenotype.
A review of this first successful screening campaign and its output will be presented along with lessons learned and future plans for the phenotypic approach to identify novel compounds suitable for pre-clinical validation as HTT lowering agents.

Disclosures: O. Lazari: A. Employment/Salary (full or part-time):; Charles River. **A. Mukonoweshuro:** A. Employment/Salary (full or part-time):; Charles River. **B. Nancolas:** A.

Employment/Salary (full or part-time):; Charles River. **C. Turnbull:** A. Employment/Salary (full or part-time):; Charles River. **I. Gowers:** A. Employment/Salary (full or part-time):; Charles River. **M. Iovino:** A. Employment/Salary (full or part-time):; Charles River. **C. Paule:** A. Employment/Salary (full or part-time):; Charles River. **R. Jarvis:** A. Employment/Salary (full or part-time):; Charles River. **P. Breccia:** A. Employment/Salary (full or part-time):; Charles River. **T. Ladduwahetty:** A. Employment/Salary (full or part-time):; Charles River. **G. McAllister:** A. Employment/Salary (full or part-time):; Charles River. **E. Doherty:** A. Employment/Salary (full or part-time):; CHDI. **J.A. Bard:** A. Employment/Salary (full or part-time):; CHDI. **D. Macdonald:** A. Employment/Salary (full or part-time):; CHDI. **A. Howard:** A. Employment/Salary (full or part-time):; CHDI. **C. Dominguez:** A. Employment/Salary (full or part-time):; CHDI. **I. Munoz-Sanjuan:** A. Employment/Salary (full or part-time):; CHDI.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.18/X24

Topic: C.04. Movement Disorders

Support: IGN14SC01

Title: Altered exocytosis of synaptic vesicles and Ca^{2+} influx in cortical neurons in a Huntington's disease mouse model studied with real-time imaging of single presynaptic terminals

Authors: *C. YU[1], S. CHEN[1], L. RONG[1], M. ZHANG[1], X. QIN[2], H. PARK[1,2]
[1]Div. of Life Sci., [2]Dept. of Physics, Hong Kong Univ. of Sci. and Technol., Hong Kong, Hong Kong

Abstract: Huntington’s disease (HD) is an autosomal dominant genetic disease caused by the abnormal expansion of the cytosine-adenine-guanine (CAG) repeat in mutant huntingtin gene. The neuronal loss was found initially in the striatum and cortex, later in other brain regions of HD patients. The alteration in synaptic transmission were reported before the initial neuronal loss in striatum and cortex. The detailed mechanisms of altered synaptic transmission in HD has not been fully understood yet. We investigated the input of the vulnerable corticostriatal synapses using real-time imaging at single presynaptic terminals. We measured the exocytosis of synaptic vesicles at single bouton level. We applied lipophilic FM 1-43 (a styryl dye) during electrical stimulation to label synaptic vesicles in cultured cortical neurons of Q175 knock-in (Q175) mice, which is closely relevant to human HD patients. FM de-staining showed that more synaptic vesicles in heterozygous neurons of Q175 mice were released than wild type. Consistently, electrophysiological results also showed decreased paired-pulse ratio in those cortical neurons. Therefore, we concluded that the release probability was increased in cortical neurons of heterozygotes. We further examined possible mechanisms of the increased release probability

using calcium imaging of presynaptic terminals with Cal520-AM (an ultrasensitive fluorescent Ca^{2+} probe). We found significant increase of Ca^{2+} influx during the electrical stimulation. Our results indicate that higher influx of Ca^{2+} during the stimulation may lead to increased release probability at cortical terminals. The application of BAPTA-AM (a Ca^{2+} chelator) rescued altered release probability in heterozygous cortical neurons, which further confirms our hypothesis. Our work will advance the current understanding of synaptic transmission at early stage of HD and contribute to finding therapeutic targets for HD.
*The first two authors contributed equally to this work.

Disclosures: C. Yu: None. **S. Chen:** None. **L. Rong:** None. **M. Zhang:** None. **X. Qin:** None. **H. Park:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.19/X25

Topic: C.04. Movement Disorders

Support: CHDI Foundation

NSF Grant ACI-1053575

Title: Cellular modeling of spiny projection neurons in a Huntington's disease mouse model

Authors: *H. SONG[1], J. GOODLIFFE[2], J. I. LUEBKE[3], C. M. WEAVER[1]
[1]Dept. of Mathematics and Computer Sci., Franklin & Marshall Col., Lancaster, PA; [2]Anat. & Neurosci., [3]Anat. & Neurobio., Boston Univ. Sch. of Med., Boston, MA

Abstract: Huntington’s disease (HD) is a fatal neurodegenerative disease with symptoms including involuntary movement, impaired judgment, personality change and depression. In HD, Spiny projection neurons (SPNs) of the striatum undergo structural and functional neuropathology in HD, but the mechanisms underlying this pathology remain unclear. Q175, a widely used transgenic HD mouse model, exhibits molecular phenotype changes, neuronal dysfunction, and subtle yet significant movement disorders. This study aims to model empirically characterized direct and indirect pathway SPNs (dSPNs and iSPNs respectively) in both wildtype (WT) and Q175 animals, to further our understanding of cellular changes in HD. Adapting a published multicompartment SPN model, we constructed stylized morphologies for dSPNs and iSPNs from WT and Q175 mice. These morphologies were consistent with morphometrics from digital reconstructions, demonstrating extra dendritic branching and lower spine density in Q175. Then we applied our custom-written parameter optimization method, implemented in NEURON, on compartmental conductance-based model SPNs to obtain model

parameters that minimize the difference between model output and empirical data. We fit the model to data from several neurons from WT dSPNs, WT iSPNs, Q175 dSPNs and Q175 iSPNs. Simulations showed that Q175 SPNs generally had lower membrane capacitance, reduced conductance and slower kinetics for the inward rectifying potassium channel at hyperpolarizing current injections, consistent with previous *in vitro* experimental results. Moreover, it was discovered that iSPNs were more excitable than dSPNs at depolarizing current injections, as observed empirically, due to several intrinsic properties differences of channel gating such as an increased conductance of fast sodium channel and decreased conductance of voltage dependent calcium channels. We also modeled synaptic responses (AMPA, GABA and NMDA), to examine how the morphologic and physiological differences of each neuron type might compound the reduced excitatory and increased inhibitory post-synaptic currents seen in Q175 compared to WT mice. This modeling study provides the first cellular model of SPNs in the Q175 HD mouse, predicting how intrinsic and synaptic properties contribute to observed firing patterns in HD, which in turn affect the striatal microcircuit.

Disclosures: H. Song: None. **J. Goodliffe:** None. **J.I. Luebke:** None. **C.M. Weaver:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.20/X26

Topic: C.04. Movement Disorders

Support: BMBF Grant 031A575B

Title: Intranasal administration of mesenchymal stem cells leads to improved survival and ameliorates behavioral phenotypes in the R6/2 mice of Huntington disease

Authors: *H. NGUYEN[1], L. YU-TAEGER[1], K. ARNOLD[2], L. DANIELYAN[3], A. STOLZING[2], E. SINGER[1], P. BAMBYNEK-DZIUK[1], J. STRICKER-SHAVER[1], A. NOVATI[1], O. RIESS[4]
[1]Univ. of Tuebingen, Tuebingen, Germany; [2]Fraunhofer Inst., Leipzig, Germany; [3]Dept. Cilnical Pharmacol., Univ. Hosp. of Tübingen, Tübingen, Germany; [4]Univ. of Tuebingen, Tuebingen, Germany

Abstract: Objectives: Huntington disease (HD) is a fatal neurodegenerative disease that is characterized by progressive impairment of motor and cognitive function and psychiatric disturbance. Although currently there is no treatment that can reverse the course of HD, studies using mesenchymal stem cells (MSCs) have shown considerable effect to ameliorate the behavioral phenotype and brain atrophy following intrastriatal administration in HD rodent models. However, intrastriatal delivery has certain disadvantages for the clinical use due to

invasiveness and host immune response. Recent updates have shown that intranasal administration (INA) of MSCs is a promising route for cell delivery to the brain. In the present study, we aim to evaluate INA as an alternative route for delivering MSCs in HD.
Methods: Bone marrow-derived MSCs at passage 3 were administered intranasally into 4-week-old R6/2 mice, which express N-terminal mutant huntingtin with approximately 140 CAG repeats. Body weight, food intake and survival rate were evaluated until 11 weeks of age. Locomotor activities and motor function were analyzed using the Labmaster system and rotarod tests.
Results: MSC-treated R6/2 mice showed an increased survival rate and decreased body weight loss at a later disease stage compared to R6/2 littermates only receiving PBS. Significantly reduced fine movements during the light phase were found in the MSC-treated group compared to the vehicle treated group, indicating an ameliorated sleep disturbance. Moreover, MSC-treated R6/2 mice exhibited a trend towards an improved rotarod performance.
Conclusion: Our results demonstrate significantly ameliorated phenotypes of R6/2 mice after MSCs administration via INA. This indicates that INA is an effective delivering route of MSCs to the brain for HD therapy.

Disclosures: H. Nguyen: None. **L. Yu-Taeger:** None. **K. Arnold:** None. **L. Danielyan:** None. **A. Stolzing:** None. **E. Singer:** None. **P. Bambynek-Dziuk:** None. **J. Stricker-Shaver:** None. **A. Novati:** None. **O. Riess:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.21/X27

Topic: C.04. Movement Disorders

Support: VGHTPE Grant VTA 10-A-2-1

Title: A novel antibody to monitor oligomeric mutant Huntingtin as a biomarker for Huntington’s disease

Authors: *C.-P. CHANG[1], M.-S. LIN[1], W.-C. SHEN[1], J.-J. SIEW[1], B.-W. SOONG[2], C.-M. CHEN[3], P.-H. TU[1], Y. CHERN[1]
[1]Academia Sinica, Taipei, Taiwan; [2]Neurolog. Inst., Taipei Veterans Gen. Hosp., Taipei, Taiwan; [3]Dept. of Neurol., Chang Gung Mem. Hosp., Taoyuan, Taiwan

Abstract: Huntington’s disease (HD) is a progressive and hereditary neurological disorder, which results from abnormal CAG/polyglutamine expansion (>35) in the N-terminus of *Huntingtin* (*HTT*). PolyQ expansion contributes to the formation of HTT aggregates in the brain and leads to progressive neurological symptoms (e.g. impaired motor and cognitive function,

psychological disorders, brain atrophy, and shorten life span) of HD patients. Mutant HTT (mHTT) has been reported in multiple peripheral tissues and blood cells. Besides mHTT aggregates, soluble mHTT oligomers are also toxic and have been implicated in the pathogenesis of HD. To date, no sensitive biomarker or effective treatment is currently available for HD. In the present study, we investigated whether accumulation of soluble mHTT oligomers in peripheral blood cells may serve as a sensitive and reliable biomarker for the onset and progression of HD. Using several standard methods (such as immunofluorescence staining, dot blot analysis and ELISA) and a newly developed anti-HTT antibody (Habe1) that recognized oligomers and aggregates of mHTT, we found that 1) Habe1 recognized the progressive accumulation of oligomeric and aggregate forms of mHTT in the brains and peripheral blood cells of HD mice (R6/2); 2) Habe1 recognized the oligomeric mHTT in human peripheral blood cells; 3) the level of oligomeric mHTT was altered in human peripheral blood cells during disease progression. Our results suggest that Habe1 could be a powerful tool to monitor oligomeric mHTT as a sensitive biomarker for HD progression.

Disclosures: C. Chang: None. **M. Lin:** None. **W. Shen:** None. **J. Siew:** None. **B. Soong:** None. **C. Chen:** None. **P. Tu:** None. **Y. Chern:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.22/X28

Topic: C.04. Movement Disorders

Support: CONACYT grant 220871

DGAPA IN214716

Title: Motor and visuospatial alterations correlate with white matter degeneration in Huntington´s disease

Authors: *V. GÁLVEZ, SR[1], A. CAMPOS-ROMO[2], G. RAMÍREZ GARCÍA[3], J. FERNANDEZ-RUIZ[4]
[1]Inst. de Neuroetología, Univ. Veracruzana, Xalapa-Enriquez, Mexico; [2]Unidad Periférica de Neurociencias, facultad de medicina UNAM/INNN, UNAM, Distrito Federal, Mexico; [3]Facultad de Medicina-Unidad Periférica de Neurociencias-INNyN MVS, Univ. Nacional Autónoma de México, México D.F., Mexico; [4]Univ. Nacional Autonoma Mexico, Mexico D.F., Mexico

Abstract: Introduction: The striatum is severely affected during the neurodegenerative process in Huntington's disease (HD). However, the possible white matter (WM) changes contribution to the main motor and cognitive impairments is unclear. **Objective:** to characterize WM changes in

incipient patients with HD, and to describe their correlation with the main motor and visuospatial alterations. **Method:** We recruited 22 patients with early clinical course and genetic diagnosis for HD; and 22 controls matched for sex, age and schooling. To evaluate motor performance, we used the UHDRS scale. For visuospatial performance, we used the Montreal cognitive evaluation (MoCA) and the CANTAB Stockings of Cambridge (SOC).Diffusion tensor images (DTI) were acquired by magnetic resonance, and analyzed using tract-based spatial statistics (TBSS), to describe the WM integrity and finally correlated with behavioral scales. **Results:** We found significant WM differences between controls and patients in the bilateral external capsule, forceps minor, forceps major and right superior longitudinal fasciculus. The UHDRS scores correlated with right anterior thalamic radiation, forceps minor, inferior fasciculus, right occipital front, and superior left longitudinal fasciculus. The MoCA-executive/visuospatial domain scores correlated with corpus callosum, left anterior corona radiata, and right inferior occipital frontal fasciculus. SOC performance correlated between number of movements to solve problems and preservation of right anterior corona radiata, inferior right occipital frontal fasciculus, right uncinated fasciculus and inferior fasciculus; and time to solve problems with left external capsule, forceps minor right and corpus callosum. **Conclusion:** An anatomical characterization of the changes in WM from patients with incipient HD was performed and correlated with the main motor and visuospatial alterations.

Disclosures: V. Gálvez: None. **A. Campos-Romo:** None. **G. Ramírez García:** None. **J. Fernandez-Ruiz:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.23/X29

Topic: C.04. Movement Disorders

Support: CONACYT Grant 22087

DGAPA Grant IN214716

CONACYT Grant 235703

Canadian Institutes of Health Research Foundation FDN-148418

Title: Implicit visuomotor learning impairment in Huntington's disease correlates with fronto-parietal and striatal atrophy

Authors: *I. VACA-PALOMARES[1], D. C. BRIEN[2], B. C. COE[2], J. FERNANDEZ-RUIZ[3], D. P. MUNOZ[4]
[1]Facultad de Psicología, Univ. Nacional Autónoma De México, Ciudad DE Mexico, Mexico;

[2]Ctr. for Neurosci. Studies, Queen's Univ., Kingston, ON, Canada; [3]Univ. Nacional Autonoma Mexico, Mexico D.F., Mexico; [4]Ctr. for Neurosci. Studies, Queens Univ., Kingston, ON, Canada

Abstract: Background: Implicit visuomotor learning has been studied with the predictive saccade task. In this task the metronomic temporal pattern of the targets allows the participant to predict the targets and actually start anticipating them. This task provides a reliable measure of visuomotor learning by quantifying the proportion of anticipatory saccades. Furthermore, this task is sensitive to neuropathology of different neurodegenerative diseases such as Huntington's disease (HD). Goals: Investigate implicit visuomotor learning and its relationship with neurodegeneration in HD using predictive-saccade task and structural Magnetic Resonance Imaging. Methods: 22 HD patients with molecular diagnosis, mean age 49.6 years ± 11.4, mean disease duration 4.5 years ± 2.9; and 22 healthy volunteers (Ctrl) with mean age 49.9 years ± 10.6 performed the Prediction Task. There were two blocks with 30 trials each. Each trial started with a red central fixation dot. After a small random fixation period, 15 dots appeared 10° left and right in either a left-right-left-right or a right-left-right-left pattern. In the Predictive block participants had to generate saccades toward a small target that alternated at a constant Inter stimulus interval (ISI) of 1000ms between two fixed locations on the horizontal meridian. In the Reactive block the same target alternating between the same two fixed locations, but one of the five ISIs (500, 750, 1000, 1250, 1500ms) was randomly used for each target step. Mean saccadic reaction time (SRT), amplitude and peak velocity were calculated for saccades within both Predictive and Reactive blocks. The study included a T1 3D volume and Diffusion-weighted imaging for both HD and Ctrl. For statistical analysis we conducted Mixed-design ANOVAs (HD/Ctrl x predictive/reactive). T1 and diffusion images were analyzed through the Voxel-based morphometry and Tract-based spatial statistic methodologies respectively. Results: The main finding showed that HD had a significant SRT increase in the predictive block ($F_{(1,43)}$=21.481, p=0.00). Furthermore, these SRTs correlated with grey-matter degeneration in right putamen, left caudate, left inferior parietal lobe and right frontal lobe Brodmann area 45. Conclusions: Our findings suggest that HD patients have implicit visuomotor learning deficits manifested as difficulties to generate anticipatory saccades in the predictive block. These deficits could be explained by fronto-parietal and striatal degeneration. The study of these processes in neurological populations could be useful to generate markers of the progression of cognitive deficits.

Disclosures: I. Vaca-Palomares: None. **D.C. Brien:** None. **B.C. Coe:** None. **J. Fernandez-Ruiz:** None. **D.P. Munoz:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.24/X30

Topic: C.04. Movement Disorders

Title: Cortical hyperactivity of neuronal microcircuits in a presymptomatic mouse model of Huntington's disease is re-balanced by metformin treatment

Authors: *I. ARNOUX[1], H. WATARI[1], M. WILLAM[2], P. NARAYAN DEY[3], S. SCHWEIGER[2], A. METHNER[3], A. STROH[1]
[1]Res. Group Mol. Imaging and Optogenetics, Inst. of Microscopic Anat. and Neurobio., Mainz, Germany; [2]Inst. of Human Genet., Mainz, Germany; [3]Dept. of Neurol. Johannes Gutenberg-University, Mainz, Germany

Abstract: Huntington's disease (HD) is an autosomal dominantly inherited neurodegenerative disorder caused by a poly-glutamine stretch in the protein huntingtin. The mutation confers toxic gains of function of the protein which generates intranuclear and cytoplasmic inclusions and ultimately results in neurodegeneration. Patients suffer from cognitive, psychiatric and motor disturbances, leading to premature death. It becomes more and more apparent that the late-onset clinical phase of HD is preceded by an extended pre-symptomatic phase with subtle molecular and clinical peculiarities. Here, we asked whether cortical network function is already impaired at the presymptomatic stage of the disease prior to neurodegeneration and motor symptoms, employing a network-centered view on early stages HD. We explored activity patterns of intact cortical microcircuits by employing *in vivo* two-photon Ca^{2+} imaging in layer 2/3 of visual cortex of mouse model of HD (HD150 mice; expressing expanded Htt with 150 glutamine repeats) at 10-15 weeks of age, long before the onset of motor symptoms and appearance of huntingtin aggregates. We identified an early pattern of circuit dysregulation characterized by an overall increase of activity, an enhanced synchronicity and a unique functional subgroup of hyperactive neurons. Exploring the potential cause of cortical dysregulation, we found that striatal but not cortical tissue exhibited increased mitochondrial respiration. Interestingly, treating presymptomatic HD150 mice with metformin rebalanced striatal mitochondrial respiration and restored physiological activity patterns of cortical microcircuit pointing to a striatal pathology behind the cortical hyperactivity. Our results suggest that metformin treatment at this very early stage might ameliorate both cellular and network dysregulations, representing a promising novel treatment strategy.

Disclosures: I. Arnoux: None. **H. Watari:** None. **M. Willam:** None. **P. Narayan Dey:** None. **S. Schweiger:** None. **A. Methner:** None. **A. Stroh:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.25/X31

Topic: C.04. Movement Disorders

Support: SAF2016-80573-R

Title: Increased translation contributes to neuronal dysfunction in Huntington's disease

Authors: J. CREUS-MUNCUNILL[1,2,3], M. GARCIA-FORN[1,2,3], R. BADILLOS[1,2,3], M. MASANA[1,2,3], J. ALBERCH[1,2,3], *E. PEREZ-NAVARRO[1,2,3]
[1]Dept. Biomed. Sciences, Univ. of Barcelona, Barcelona, Spain; [2]Inst. d'Investigacions Biomèdiques August Pi i Sunyer (IDIBAPS), Barcelona, Spain; [3]Ctr. de Investigación Biomédica en Red sobre Enfermedades Neurodegenerativas (CIBERNED), Madrid, Spain

Abstract: Huntington's disease (HD) is a neurodegenerative disease caused by a CAG repeat expansion in the exon-1 of the huntingtin gene. Striatal projection neurons are the main affected population, but the molecular mechanisms that account for this specific vulnerability are not fully characterized. Mutant huntingtin expression disrupts the normal signaling of the mTOR/Akt pathway, and one of the most important functions of mTOR is the regulation of protein translation. Since dysregulation of mTOR pathway, and consequently protein synthesis, has been proposed as a pathogenic mechanism in different neurodegenerative and neurological disorders, here we sought to analyze whether protein translation is altered in HD. For that, we focused on the study of 4E-BP1, a binding protein substarte of mTOR, that inhibits protein translation and whose function is lost upon phosphorylation. We analyzed the phosphorylation status of 4E-BP1 in the striatum of HD mouse models and affected patients. We found that 4E-BP1 is inactivated in HD by increased phosphorylation leading to an increased formation of the eIF4F complex. Accordingly, we detected aberrant *de novo* protein translation at ages when HD mice suffer from motor deficits. Interestingly, normalization of protein synthesis in R6/1 mice, a transgenic mouse model of HD, prevents motor learning deficits. Finally, we observed that genetically increasing protein translation in adult wild-type mice recapitulates HD phenotype and induces neurodegeneration. Our results seem to indicate that dysregulation of protein synthesis is a novel key event in HD pathogenesis.

Disclosures: J. Creus-Muncunill: None. **M. Garcia-Forn:** None. **R. Badillos:** None. **M. Masana:** None. **J. Alberch:** None. **E. Perez-Navarro:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.26/X32

Topic: C.04. Movement Disorders

Support: RGC GRF 14122815

Title: Dissecting the regulatory mechanism of ubiquitin E3 ligase activity and understanding the importance of such regulation in polyglutamine pathogenesis

Authors: *E. CHAN, Z. CHEN, L. LI
The Chinese Univ. of Hong Kong, Hong Kong, Hong Kong

Abstract: Polyglutamine (polyQ) diseases, including Huntington's Disease and several types of spinocerebellar ataxias, are caused by the genomic expansion of coding CAG trinucleotide sequences. This expansion leads to the production and accumulation of misfolded polyQ domain-containing disease proteins, which causes cellular dysfunction and neuronal death. As one of the principal cellular protein clearance pathways, the activity of Ubiquitin-Proteasome system (UPS) is tightly regulated to ensure efficient clearance of damaged and toxic proteins. Emerging evidence demonstrates that UPS plays a crucial role in the pathogenesis of polyQ diseases. Ubiquitin (Ub) E3 ligase label proteins for proteasomal clearance by catalyzing the transfer of the Ub tag to protein substrates. We recently identified an E3 ligase that modulates expanded polyQ-induced neurodegeneration in both mammalian and *Drosophila* disease models. We further showed that it promotes polyubiquitination and degradation of expanded polyQ protein. Intriguingly, we found that exocyst complex protein 70 (Exo70/EXOC7) modulates expanded polyQ protein level and toxicity in an opposite manner to E3 ligase. Our data suggest that Exo70/EXOC7 exerts, at least in part, its polyQ-modifying effect by regulating the function of E3 ligase. In summary, this study allows us to better define the role of protein ubiquitination in polyQ pathogenesis and understand how E3 ligase activity can be regulated by Exo70/EXOC7.

Disclosures: E. Chan: None. **Z. Chen:** None. **L. Li:** None.

Poster

388. Cell Biology of Huntington's Disease I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 388.27/X33

Topic: C.04. Movement Disorders

Support: NIH grant 5P30GM103398-02 (pilot project)

NIH grant 2P20GM103432

NIH grant R01NS079450

HDSA Donald A. King Summer Fellowship

Title: Latent toxoplasma gondii infection promotes neurodegeneration and increases soluble mutant huntingtin levels in the YAC128 mouse model of Huntington's disease

Authors: *D. DONLEY[1], *D. DONLEY[1], T. JENKINS[2], M. REALING[2], V. CHOPRA[4], S. HERSH[4], J. GIGLEY[2], J. H. FOX[3]
[1]Neurosci. Program, [3]Vet. Sci., [2]Univ. of Wyoming, Laramie, WY; [4]Mass Gen., Charleston, MA

Abstract: Huntington disease (HD) is a fatal neurodegenerative disorder caused by a dominant CAG repeat expansion in the huntingtin gene that manifests with motor, cognitive and psychiatric symptoms. The kynurenine pathway of tryptophan metabolism is a neuroinflammatory pathway implicated in HD pathogenesis. Activity of the first enzyme in this pathway, indoleamine-2,3-dioxygenase (IDO), is increased in HD mouse and human brain. IDO activation also occurs as part of the immune response to the prevalent neuroinvasive human pathogen, *Toxoplasma gondii (T. gondii)* and is required for prolonged control of the infection. Therefore, latent infection activates an HD-associated pathway and may exacerbate disease. We previously reported that N171-82Q HD mice have an altered response to acute *T. gondii* infection (Donley et al. 2016). However, the effect of latent *T. gondii* infection, characterized by intracellular parasite cysts and inflammation in brain, on HD progression is not known. Here we investigated the effects of latent *T. gondii* infection in YAC128 HD mice. Wild-type and HD mice were orally infected at 2 months of age with *T. gondii* or vehicle. Behavior was monitored longitudinally and mice were sacrificed at 12-months of age. Both infection and HD increased IDO activity but there was no statistically significant difference in IDO enzyme activity in infected HD mice. Interestingly, infection increased soluble mutant huntingtin levels in HD mice compared to HD mice without infection. In addition, brain weight of *T. gondii*-infected HD mice was significantly decreased as compared to both non-infected HD mice and wild-type mice with infection. These results are consistent with HD potentiation by latent *T. gondii* infection in the YAC128 mouse model.

Disclosures: D. Donley: None. **T. Jenkins:** None. **M. Reading:** None. **V. Chopra:** None. **S. Hersh:** None. **J. Gigley:** None. **J.H. Fox:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.01/Y1

Topic: C.04. Movement Disorders

Support: MJD Foundation Grant

NHMRC Project Grant 1069235

MQ Uni Research Development Grant

Title: Drug testing in a transgenic zebrafish model of neurodegenerative disease identifies drugs that can remove toxic human proteins

Authors: *A. LAIRD[1], M. WATCHON[2], K. YUAN[1], N. J. COLE[1], G. NICHOLSON[3]
[1]Dept. of Biomed. Sci., Macquarie Univ., Sydney, Australia; [2]Univ. of Sydney, Sydney, Australia; [3]ANZAC Res. Inst., Sydney, Australia

Abstract: Spinocerebellar ataxia-3 (SCA3), also known as Machado-Joseph disease, is a fatal neurodegenerative disease that affects neurons of the brain and spinal cord causing impaired movement. SCA3 is caused by inheritance of a long trinucleotide (CAG) repeat region within the *ATXN3* gene. This CAG repeat region encodes for a polyglutamine (polyQ) tract that usually contains less than 43 glutamines (43Q), but in SCA3 patients contains more than 60Q. We have developed the first transgenic zebrafish model of SCA3, zebrafish that express the human ataxin-3 protein containing either 23Q (wild-type) or 84Q (SCA3-causing). The ataxin-3 84Q zebrafish develop impaired swimming ability as early as 6 days old. We have tested the effects of various drug and small compound treatments on these zebrafish and have identified drugs that improve the swimming of the zebrafish. Immunoblot analysis of protein lysates extracted from the SCA3 zebrafish reveal that the drug treatments have produced removal of the human ataxin-3 protein through activity of the autophagy quality control pathway. Development of an ELISA assay to quantify levels of human ataxin-3 protein confirmed the complete removal of human ataxin-3 by one compound. These findings confirm that our transgenic zebrafish model of SCA3 is a useful model of the human disease. We have also identified candidates for further exploration for the treatment of SCA3, and perhaps other neurodegenerative diseases as well.

Disclosures: A. Laird: None. **M. Watchon:** None. **K. Yuan:** None. **N.J. Cole:** None. **G. Nicholson:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.02/Y2

Topic: C.04. Movement Disorders

Support: Kennedy's Disease Association

Title: Targeting the acetyltransferase activity of Tip60 abrogates aspects of spinal and bulbar muscular atrophy pathology

Authors: *H. L. MONTIE[1], D. ESTRADA[1], C. MEZES[1], A. LUKASIK[1], E. HEINE[1], D. EXLER[1], Y. LIU[2], G. ZHENG[3], F. DEKKER[4], D. E. MERRY[2]
[1]Bio-Medical Sci., Philadelphia Col. of Osteo. Med., Philadelphia, PA; [2]Dept. of Biochem. and

Mol. Biol., Thomas Jefferson Univ., Philadelphia, PA; [3]The Univ. of Georgia, Athens, GA; [4]Univ. of Groningen, Groningen, Netherlands

Abstract: Spinal and Bulbar Muscular Atrophy (SBMA; Kennedy's Disease) is an adult-onset, slowly-progressive, X-linked neuromuscular disease. SBMA results from a CAG repeat expansion in the coding region of the androgen receptor (AR) gene, leading to a polyglutamine (polyQ) repeat expansion in the amino-terminal domain of the AR protein. Patients present with weakness and atrophy of muscles of the limbs, mouth, and throat. These symptoms result from the loss of lower motor neurons and skeletal muscle atrophy. A histopathological hallmark of SBMA, as well as other polyQ-repeat diseases, is the presence of intranuclear inclusions of the polyQ-expanded AR protein. To date, there is no curative treatment or effective therapy for SBMA. We have previously shown that hormone-dependent AR acetylation at K630/632/633 is a major modifier of polyQ-expanded AR toxicity and inclusion formation. Tip60 (Tat-interactive protein, 60kDa) functions as a histone acetyltransferase (HAT) and has been shown to directly acetylate the AR at K630/632/633. We are studying the role of Tip60 in SBMA by reducing its protein level and by pharmacologically inhibiting its acetyltransferase activity in various cell models of SBMA. We have found that knock-down of Tip60 protein, as well as pharmacologic inhibition of Tip60, in a PC12 cell model of SBMA, reduces the formation of nuclear inclusions of polyQ-expanded AR. Moreover, pharmacologic inhibition of Tip60 reduces toxicity in both myoblast and primary motor neuron models of SBMA. Mechanistic studies are underway and *in vivo* validation studies are planned for the future. The goal of these studies is to identify whether Tip60 is a viable therapeutic target for the treatment of SBMA.

Disclosures: H.L. Montie: None. **D. Estrada:** None. **C. Mezes:** None. **A. Lukasik:** None. **E. Heine:** None. **D. Exler:** None. **Y. Liu:** None. **G. Zheng:** None. **F. Dekker:** None. **D.E. Merry:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.03/Y3

Topic: C.04. Movement Disorders

Support: Ministry of Science and Technology, Taiwan (NSC101-2321-B-001-017)

Institute of Biomedical Sciences, Academia Sinica, Taiwan

Title: Reduced cytoplasmic MBNL1 is an early event in a brain-specific mouse model of myotonic dystrophy

Authors: *P.-Y. WANG[1,2], Y.-M. LIN[1], L.-H. WANG[1], T.-Y. KUO[1], S.-J. CHENG[1,3], G.-S. WANG[1]
[1]Inst. of Biomed. Sci., Taipei, Taiwan; [2]Program in Mol. Medicine, Natl. Yang-Ming Univ. and Academia Sinica, Taipei,, Taiwan; [3]Neurosci. Program of Academia Sinica,, Taipei, Taiwan

Abstract: Myotonic dystrophy type 1 (DM1) is caused by an expansion of CTG repeats in the 3' untranslated region (UTR) of the dystrophia myotonia protein kinase (DMPK) gene. Cognitive impairment associated with structural change in the brain is prevalent in DM1. How this histopathological abnormality during disease progression develops remains elusive. Nuclear accumulation of mutant DMPK mRNA containing expanded CUG RNA disrupting the cytoplasmic and nuclear activities of muscleblind-like (MBNL) protein has been implicated in DM1 neural pathogenesis. The association between MBNL dysfunction and morphological changes has not been investigated. We generated a mouse model for postnatal expression of expanded CUG RNA in the brain that recapitulates the features of the DM1 brain, including the formation of nuclear RNA and MBNL foci, learning disability, brain atrophy and misregulated alternative splicing. Characterization of the pathological abnormalities by a time-course study revealed that hippocampus-related learning and synaptic potentiation were impaired before structural changes in the brain, followed by brain atrophy associated with progressive reduction of axon and dendrite integrity. Moreover, cytoplasmic MBNL1 distribution on dendrites decreased before dendrite degeneration, whereas reduced MBNL2 expression and altered MBNL-regulated alternative splicing was evident after degeneration. These results suggest that the expression of expanded CUG RNA in the DM1 brain results in neurodegenerative processes, with reduced cytoplasmic MBNL1 as an early event response to expanded CUG RNA.

Disclosures: P. Wang: None. **Y. Lin:** None. **L. Wang:** None. **T. Kuo:** None. **S. Cheng:** None. **G. Wang:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.04/Y4

Topic: C.04. Movement Disorders

Support: National Ataxia Foundation

FEDER (COMPETE)

Portuguese Foundation for Science and Technology

COMPETE:POCI-01-0145-FEDER-007440

EXPL/NEU-NMC/0331/2012

SFRH/BPD/66705/2009

PTDC/NEU-NMC/0084/2014

Title: Non-invasive and allele-specific silencing of mutant ataxin-3 alleviates neuropathology and motor deficits of Machado-Joseph disease

Authors: R. NOBRE[1,2], J. SARAIVA[1], C. FUSCO[1], S. PAIXÃO[1], M. SANTANA[1,2], M. ESTEVES[3], *L. PEREIRA DE ALMEIDA[1,4]
[1]CNC - Ctr. For Neurosci. | Univ. of Coimbra, Coimbra, Portugal; [2]Inst. for Inter-disciplinary Research, Univ. of Coimbra, Portugal, Coimbra, Portugal; [3]Neurol. Department, Gene Therapy Center, Univ. of Massachusetts Med. School, MA, USA, Worcester, MA; [4]Fac. of Pharmacy, Univ. of Coimbra, Portugal, Coimbra, Portugal

Abstract: Machado-Joseph disease (MJD) is the most common dominantly-inherited ataxia. It is associated with the expansion of a (CAG)n tract in the coding region of the MJD1/ATXN3 gene. This abnormal over-repetition is translated into an expanded polyglutamine tract within ataxin-3, conferring toxic properties to this protein and resulting in severe clinical features. Although there is no cure, several preclinical studies have demonstrated that silencing mutant ataxin-3 expression using RNA interference (RNAi) is a promising therapeutic approach. In particular, our group showed that intracranial injection of viral vectors targeting mutant ataxin-3 significantly decreases the severity of the neuropathological abnormalities in rodent models of MJD. However, this is an invasive procedure, which is associated with potential adverse effects and a limited vector distribution in the brain.
The aim of the present study was to develop an adeno-associated virus (AAV)-based system that enables: delivery of RNA interference-based treatments to the brain, the specific silencing of mutant ataxin-3 (mutATAX3) and alleviation of the disease by intravenous (iv) injection.
For that, we used adeno-associated viral vector serotype 9 (AAV9), a vector that has a remarkable ability to bypass the blood-brain barrier (BBB) and transduce the central nervous system of mammals. AAV9 vectors encoding an artificial microRNA that targets the mutant form of ataxin-3 mRNA (AAV9-mirATAX3) were firstly generated. Its efficacy and specificity were firstly confirmed in neuronal cell models and viral-based mouse models and the therapeutic potential was then tested in a severely impaired transgenic mouse model of MJD. Mice were intravenously injected at postnatal day one (PN1); were submitted to behavioral tests at 3 different ages and sacrificed at PN95.
AAV9-mirATAX3 vectors efficiently spread throughout the brain, transducing regions affected in MJD, such as the striatum, cerebellum, brainstem and spinal cord. AAV9-mirATAX3's treatment reduced the number of mutATAX3 aggregates and the cerebellar neuropathology, and treated animals showed a better performance in all behavioral tests. Overall, this study provides compelling evidence that a single iv injection of AAV9-mirATAX3 at PN1 is able to transpose the BBB, silence mutATAX3 and alleviate MJD motor phenotype.
To our knowledge, this is the first time that a non-invasive iv administration of rAAV9 vectors had significant impact on motor deficits of a polyglutamine disorder.

Disclosures: **R. Nobre:** None. **J. Saraiva:** None. **C. Fusco:** None. **S. Paixão:** None. **M. Santana:** None. **M. Esteves:** None. **L. Pereira de Almeida:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.05/Y5

Topic: C.04. Movement Disorders

Support: Doctoral Program in Molecular Medicine, University of Eastern Finland

VTR 5772816, Kuopio University Hospital

Title: Involvement of the frontotemporal dementia gene C9ORF72 in proteasome- and autophagy- mediated protein degradation pathways

Authors: *S. S. LESKELÄ[1], M. A. TAKALO[2,1], J. LIST[1], M. CARTRÓ FONT[1], H. SOININEN[3,4], A. M. REMES[3,4], M. HILTUNEN[2,4], A. HAAPASALO[1,4]
[1]A.I. Virtanen Inst. for Mol. Sci., [2]Inst. of Biomedicine, [3]Inst. of Clin. Med. - Neurol., Univ. of Eastern Finland, Kuopio, Finland; [4]NeuroCenter, Neurol., Kuopio Univ. Hosp., Kuopio, Finland

Abstract: Frontotemporal dementia (FTD) is the second most common cause of dementia within the working-age population. Molecular pathogenesis of FTD is suggested to involve impairment of protein degradation pathways, including the ubiquitin-proteasome system (UPS) and autophagy, leading to formation of intracellular inclusions containing e.g. TAR DNA-binding protein 43, Fused in Sarcoma or ubiquilin proteins. Here, we have studied the involvement of *C9ORF72*, the major genetic contributor to FTD and amyotrophic lateral sclerosis (ALS), in these protein degradation pathways. We have utilized overexpression of C9orf72 protein isoforms A and B and modulation of UPS or autophagy-mediated degradation pathways by the proteasomal inhibitor lactacystin or serum starvation with or without Bafilomycin A1, an inhibitor of autophagosome maturation, in N2a mouse neuroblastoma cells. We observed that the levels of both C9orf72 isoforms significantly increased after proteasomal inhibition, suggesting that C9orf72 protein levels are regulated by proteasomal degradation. There were no signs of C9orf72 aggregation in the cells under these conditions. Induction of autophagy led to a significant decrease in the levels of both C9orf72 isoforms. Co-immunoprecipitation studies did not indicate interaction between C9orf72 and autophagy marker proteins SQSTM1/p62 or LC3B. Despite some recent studies, which have suggested the involvement of C9orf72 proteins in autophagy initiation, we did not observe changes in the induction of autophagy in cells overexpressing either of the C9orf72 isoforms compared to control cells, as indicated by unaltered LC3BII/LC3BI ratio. Our results confirm that C9orf72 proteins are linked to UPS and

autophagy pathways. Ongoing studies will provide further insights on the mechanistic role of C9orf72 proteins in the regulation of these protein degradation mechanisms.

Disclosures: S.S. Leskelä: None. **M.A. Takalo:** None. **J. List:** None. **M. Cartró Font:** None. **H. Soininen:** None. **A.M. Remes:** None. **M. Hiltunen:** None. **A. Haapasalo:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.06/Y6

Topic: C.04. Movement Disorders

Support: NIH Grant R01NS096176

NIH Grant R01NS097231

Title: The ALS/FTD associated protein C9ORF72 forms a complex with SMCR8 to regulate ULK1 and play a dual role in autophagy

Authors: *C. LIANG[1,2], M. YANG[1], K. SWAMINATHAN[3], F. LAI[4], R. SHIEKHATTAR[4], J.-F. CHEN[2]
[1]Univ. of Georgia, Athens, GA; [2]USC, Los Angeles, CA; [3]Natl. Univ. of Singapore, Singapore, Singapore; [4]Univ. of Miami Miller Sch. of Med., Miami, FL

Abstract: The intronic GGGGCC hexanucleotide repeat expansion in chromosome 9 open reading frame 72 (C9ORF72) is the most common genetic cause of both frontotemporal dementia (FTD) and amyotrophic lateral sclerosis (ALS). Reduced C9orf72 expression levels have been reported in C9FTD/ALS patients, however the normal function of C9orf72 remains largely unknown. We report that C9ORF72 is a component of a multiprotein complex containing SMCR8, WDR41, and ATG101. Both C9ORF72 and SMCR8 have been predicted to contain DENN domains. We created Smcr8 knockout mice and found that they exhibit similar splenomegaly and lymphadenopathy as C9orf72 knockout mice, suggesting that the two proteins may have similar functions. Although both C9ORF72 and SMCR8 have been implicated in autophagy, experimental evidence of their functions is lacking. We found that Smcr8 knockout cells exhibit impaired autophagy induction, which is similarly observed in C9orf72 knockdown cells. Mechanistically, C9ORF72/SMCR8 interacts with the key autophagy initiation ULK1/FIP200/ATG13 complex and the interaction is enhanced under amino acid starvation conditions. Smcr8 regulates the expression of the ULK1 complex and has an opposite role in regulating the activity of ULK1 compared with C9orf72. In addition to modulating autophagy induction, the complex also regulates later stages of autophagy. Whereas autophagic flux is increased in C9orf72 knockdown cells, depletion of Smcr8 results in a reduced flux with an

abnormal expression of lysosomal proteases. Thus, C9ORF72 and SMCR8 have similar functions in modulating autophagy induction by interacting with the ULK1 complex and play distinct roles in regulating late steps of autophagy.

Disclosures: C. Liang: None. **M. Yang:** None. **K. Swaminathan:** None. **F. Lai:** None. **R. Shiekhattar:** None. **J. Chen:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.07/Y7

Topic: C.04. Movement Disorders

Support: KAKENHI（26293207）

Title: Induction of autophagy enhances degradation of causative proteins in cellular models of neurodegenerative diseases

Authors: *H. ADACHI, Z. HUANG, K. OKADA, K. OHNARI, T. HASHIMOTO, T. TOYOTA, Y. IWANAKA
Dept of Neurol., Univ. of Occup. and Envrn. Hlth., Kitakyushu-Shi, Japan

Abstract: Neurodegenerative diseases share the common morphological characteristic of the deposition of abnormal proteins in the nervous system, including neurons. In chronic neurodegenerative diseases such as polyglutamine (polyQ) and Alzheimer disease (AD), commonly observed phenotypes include the abnormal accumulation of disease-causing proteins and the formation of nuclear and cytoplasmic inclusions. Under pathologic conditions, the accumulated level of such misfolded and toxic proteins may exceed the protective ability of the proteolytic machinery; the inability to either maintain misfolded proteins in a soluble form or degrade them results in their accumulation and the formation of inclusions. Autophagy refers to self-phagocytosis and is a process by which cells remove a large amount of proteins with long half-lives as well as damaged organelles. The mechanism of autophagy involves double-membraned autophagic vacuoles encapsulating the cytoplasm and organelles, and their fusion with lysosomes, resulting in the degradation of intracellular components. We examined the effects of the kaempferol in cultured cell models of neurodegenerative diseases. Cells were transfected using Lipofectamine 2000 with plasmids encoding mutant androgen receptor and ataxin-1. Kaempferol decreased the expression of both causative proteins in the neuronal cell models. The expression of the autophagic marker LC-3 II was significantly elevated in the cells after treatment with kaempferol. Kaempferol also decreased the expression of p62. These findings demonstrated that kaempferol induced autophagosome formation and enhanced the preferential degradation of the disease-causative proteins.

Disclosures: **H. Adachi:** None. **Z. Huang:** None. **K. Okada:** None. **K. Ohnari:** None. **T. Hashimoto:** None. **T. Toyota:** None. **Y. Iwanaka:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.08/Y8

Topic: C.04. Movement Disorders

Support: MSU-Mankato Biology Department

Title: PolyQ protein FAM171B expression and localization in mouse brain

Authors: Q. H. TRAN[1], A. K. SUDASINGHE[2], B. JONES[3], D. SHARLIN[3], *G. M. GOELLNER[4]
[1]Biol. Sci., [2]biology department, [3]Minnesota State Univ., Mankato, MN; [4]Biol., Minnesota State Univ- Mankato, Mankato, MN

Abstract: Polyglutamine (polyQ) diseases are inherited fatal neurodegenerative disorders caused by expansion of trinucleotide cytosine-adenine-guanine (CAG) repeats, encoding abnormally long glutamine tracts in respective disease proteins. Currently, there are nine polyQ diseases- including Huntington's disease and a number of Spinocerebellar ataxias. Interestingly, expanded polyQ proteins are prone to aggregate, and this aggregation may underlie neurodegeneration. In this study, we investigate the expression and localization of FAM171B (a novel polyQ protein) in the brain. Western blotting reveals that FAM171B protein is indeed expressed in the developing and adult mouse brain. Furthermore, *in situ* hybridization and immunohistochemical analyses suggests widespread localization of FAM171B to many brain regions- with pronounced expression found in the hippocampus, cerebellar Purkinje cells, and cerebral cortex. As a novel polyQ protein that is expressed in the brain, our observations suggest that FAM171B can be considered a candidate gene for an as yet molecularly uncharacterized neurodegenerative disease.

Disclosures: **Q.H. Tran:** None. **A.K. Sudasinghe:** None. **B. Jones:** None. **D. Sharlin:** None. **G.M. Goellner:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.09/Y9

Topic: C.04. Movement Disorders

Title: Ataxin-2 in stress granules regulate nucleocytoplasmic transport in als

Authors: *K. ZHANG[1], J. G. DAIGLE[2], K. M. CUNNINGHAM[3], A. N. COYNE[4], J. C. GRIMA[5], T. E. LLOYD[6], J. D. ROTHSTEIN[4]
[1]Dept. of Neurol., [2]Neurology/Neurosurgery, Brain Sci. Inst., [3]CMM, [4]Brain Sci. Inst., [5]Neurosci., [6]Neurol., Johns Hopkins University, Sch. of Med., Baltimore, MD

Abstract: Nucleocytoplasmic transport defects have been identified as a key pathogenic mechanism in C9orf72-mediated amyotrophic lateral sclerosis (ALS), the most common familial ALS. Recently, they have been also implicated in Huntington's disease and tauopathy and are suggested to be a common pathology underlying cytoplasmic protein aggregation diseases. However, how does cytoplasmic protein aggregations disrupt nucleocytoplasmic transport is unclear. In an effort to understand whether Ataxin-2, an ALS risk factor and also stress granule protein, modulates nucleocytoplasmic transport, we serendipitously identified that Ataxin-2 physically and genetically interacts with Ran GTPase (Ran), a key regulator of nucleocytoplasmic transport. Furthermore, we found that Ran is mislocalized into the stress granules when cells are under stress, potentially through Ataxin-2. Moreover, cells treated with stressors exhibit nucleocytoplasmic transport defects that can be rescued by inhibiting stress granule assembly, suggesting that stress granules regulate nucleocytoplasmic transport. As stress granules also play an important role in ALS pathogenesis, we next tested whether stress granule proteins contributes to the nucleocytoplasmic transport defects observed in C9orf72-mediated ALS. Indeed, we found that molecules inhibiting stress granule assembly suppresses these defects as well as neurodegeneration in fly and/or iPS models of C9orf72-mediated ALS. Hence, our studies delineates the stress granule-nucleocytoplasmic transport pathway that plays a role in pathogenesis of C9orf72-mediated ALS and potentially other cytoplasmic protein aggregation diseases.

Disclosures: K. Zhang: None. **J.G. Daigle:** None. **K.M. Cunningham:** None. **A.N. Coyne:** None. **J.C. Grima:** None. **T.E. Lloyd:** None. **J.D. Rothstein:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.10/Y10

Topic: C.04. Movement Disorders

Support: University of Pennsylvania Orphan Disease Center

Applied Genetic Technologies Corp

Title: Peripheral nervous system pathology in a mouse model of adrenomyeloneuropathy

Authors: *F. LAHEJI, Y. GONG, A. BERENSON, R. KOK, F. EICHLER
Massachusetts Gen. Hosp., Boston, MA

Abstract: X-linked adrenoleukodystrophy (X-ALD) is a devastating neurological disorder caused by mutations in the ABCD1 gene that encodes a peroxisomal ATP-binding cassette (ABC) transporter. The mouse model of X-ALD with ABCD1 deficiency develops a phenotype similar to adrenomyeloneuropathy (AMN), the most common phenotype of X-ALD which manifests as spinal cord axonopathy and peripheral neuropathy. We previously observed mechanical hypersensitivity in the ABCD1-deficient mouse as early as 8 months of age, as detected by Von Frey filament testing. Here, we report various pathological parameters of the peripheral nervous system in an attempt to explain this hyperpathia. Density of PGP9.5-positive nerve fibers in the AMN mouse footpad was diminished at 14 months of age, though not at 8 months of age. Myelination in the sciatic nerve was also diminished at old age. No difference in the percentage of small-size nociceptive neurons in dorsal root ganglia (DRG) as determined by peripherin/neurofilament costaining was seen at either young (1 month) or old age (13 months). DRG neurons from AMN mice displayed a 56% increase in neurite branching at 1 month of age, although neurite length did not differ from wild type mice. The changes in footpad nerve fiber density and sciatic nerve myelination in older mice contribute to the understanding of AMN disease progression, but do not explain the hypersensitivity detected in younger mice. Early changes in DRG neurite branching suggest that developmental morphological changes in DRG neurons may contribute to the observed hyperpathia. Further investigation of neuropathological changes in young mice is underway.

Disclosures: F. Laheji: None. **Y. Gong:** None. **A. Berenson:** None. **R. Kok:** None. **F. Eichler:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.11/Y11

Topic: C.04. Movement Disorders

Support: MIUR (SIR project RBSI14B1Z1)

Title: Down regulating mGluR5 in the $SOD1^{G93A}$ mouse model of ALS reduces the reactive phenotype of ex-vivo cultured spinal cord astrocytes

Authors: *C. USAI[1], F. PROVENZANO[2], E. GALLIA[2], M. MILANESE[2,3], T. BONIFACINO[2], G. BONANNO[2,3]
[1]Natl. Rese Council, Genova, Italy; [2]2Department of Pharmacy, Unit of Pharmacol. and Toxicology, Univ. of Genoa, 16132 Genoa, Italy, Genoa, Italy; [3]Ctr. of Excellence for Biomed. Research, Univ. of Genoa, 16148 Genoa, Italy, Genoa, Italy

Abstract: Amyotrophic lateral sclerosis (ALS) is a lethal neurodegenerative disease characterized by a selective death of upper and lower motor neurons (MNs). Although the etiopathogenesis is not completely understood, *in-vitro* and *in-vivo* studies demonstrated that damage within MNs is sustained by the degeneration of non-neuronal cells such as microglia and astrocytes (1). Group I metabotropic glutamate receptors (mGluR1, mGluR5) likely play a role in ALS, since they are over-expressed and functionally altered in different experimental model of ALS (2,3,4). We demonstrated that knocking-down mGluR1 or mGluR5 significantly prolongs survival and ameliorates the clinical progression in the $SOD1^{G93A}$ mouse model of ALS (5,6). The aim of this work is to investigate the effects of mGluR5 knocking-down on the reactive phenotype of astrocytes in ALS. We used here spinal cord astrocyte cell cultures from adult $SOD1^{G93A}$ mice and $SOD1^{G93A}mGluR5^{+/-}$ mice, heterozygous for mGluR5, and WT mice. Experiments with the FURA-2 dye showed a significantly higher cytosolic calcium concentration ($[Ca^{2+}]_i$) in $SOD1^{G93A}$ than in WT mice, both under basal condition and after exposure to the Group I mGluRs agonist 3,5-DHPG (30 μM). mGluR5 knocking-down significantly reduced the excessive $[Ca^{2+}]_i$. Confocal microscopy revealed that the astrogliosis markers GFAP, vimentin and S100β were more expressed in $SOD1^{G93A}$ respect to WT mice and decreased in $SOD1^{G93A}mGluR5^{+/-}$ mice. The same was true for the expression of the authophagy activation marker LC3. Of note, mGluR5 knocking-down also translates in a significant lower presence of misfolded-SOD1 protein when comparing $SOD1^{G93A}mGluR5^{+/-}$ and $SOD1^{G93A}$ mice. To conclude, a lower constitutive level of mGluR5 had a positive impact in $SOD1^{G93A}$ mouse astrocytes, supporting the idea that mGluR5 may be a potential pharmacological target for cell specific therapeutic approaches in ALS, aimed at preserving MNs by acting at the neighboring astroglial cell. 1. Philips and Rothstein 2014. Exp Neurol. S0014-4886; 2. Aronica et al. 2001.

Neurosc. 105:509-20; 3. Rossi et al. 2008. Cell Death Diff. 15:1691-700; 4. Giribaldi et al. 2013. Neuropharmacol. 66:253-63; 5. Milanese et al. 2014. Neurobiol. Dis. 64:48-9; 6. Bonifacino et al. 2017. Neuropharmacol., accepted.

Disclosures: C. Usai: None. **F. Provenzano:** None. **E. Gallia:** None. **M. Milanese:** None. **T. Bonifacino:** None. **G. Bonanno:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.12/Y12

Topic: C.04. Movement Disorders

Support: NIMH Grant 1R01MH106174

Brown University Dean's Emerging Areas of New Science Award

Title: Non-conventional DBS stimulation for tremor and non-tremor symptoms of ET

Authors: *S. LEE[1], W. F. ASAAD[2], S. R. JONES[3]
[1]Neurosci., [2]Neurosurg., [3]Dept. of Neurosci., Brown Univ., Providence, RI

Abstract: Essential Tremor (ET) is a movement disorder in which patients experience an involuntary intention tremor at 4-8 Hz. Tremor frequency oscillations have been widely observed during tremor, but whether there is a primary neurophysiological source of this oscillation is not fully known. For medication-refractory ET, deep brain stimulation (DBS) of the ventral intermediate nucleus of the thalamus (VIM) can be an effective treatment. However, DBS is not universally effective and may cause cerebellar-mediated side-effects such as gait imbalance, abnormal timing of motor activities, and deficits in motor learning. Here, we developed a biophysically principled computational neural model of tremor frequency oscillations in VIM, along with simulated DBS. We compared tremor oscillations generated by two candidate mechanisms: intrinsically within the thalamus and driven by cerebellar tremor frequency inputs. We compared the local fields and spiking of both mechanisms to human intraoperative data and show that both mechanisms may be plausible. We then simulated an additional source of cerebellar inputs as a model for non-tremor-frequency information and show that only the driven tremor oscillations were robust to these inputs. We simulated DBS during ongoing tremor and show that both standard and non-standard temporal patterns of DBS reduced tremor frequency oscillations in our model. We present predictions from the simulation results and suggest that improved understanding of the actions of DBS on both tremor and non-tremor activity in VIM may result in improved treatment efficacy.

Disclosures: S. Lee: None. **W.F. Asaad:** None. **S.R. Jones:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.13/Y13

Topic: C.04. Movement Disorders

Title: Implementation of a multilayer perceptron neural network for classifying deep brain stimulation in 'On' and 'Off' modes through a smartphone representing a wearable and wireless sensor application

Authors: *R. C. LEMOYNE[1], T. J. MASTROIANNI[2]
[1]Independent, Running Springs, CA; [2]Cognition Engin., Pittsburgh, PA

Abstract: Essential Tremor is a prevalent movement disorder that is characterized by postural and action tremor. In the event that medication therapy becomes intractable deep brain stimulation (DBS) of the VIM (ventralis intermedius) nucleus of the thalamus provides a viable alternative. However, the tuning process by an expert clinician is a laborious endeavor. The smartphone is equipped with an inertial sensor package comprising an accelerometer and gyroscope. A software application enables the smartphone to function as a wearable and wireless inertial sensor system for measuring the severity of Essential Tremor in a quantified methodology. The status of a subject with the DBS device in 'On' and 'Off' mode can be readily contrasted from a quantified perspective based on the recorded inertial sensor signal. The numeric attributes of the signal can be consolidated into a feature set for machine learning classification, such as through a multilayer perceptron neural network. The multilayer perceptron neural network achieves considerable classification accuracy with regards to distinguishing between a subject with Essential Tremor with the DBS device in 'On' and 'Off' mode through the inertial sensor signal recording of a smartphone functioning as a wearable and wireless sensor application.

Disclosures: R.C. LeMoyne: None. **T.J. Mastroianni:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.14/Y14

Topic: C.04. Movement Disorders

Title: Characterizing the deep brain stimulation micro-lesion effect using peripheral sensors

Authors: *S. CERNERA[1], E. OPRI[1], A. GUNDUZ[2]
[2]Biomed. Engin., [1]Univ. of Florida, Gainesville, FL

Abstract: Introduction Essential tremor (ET) is one of the most prevalent hyperkinetic adult movement disorders, affecting roughly 1% of the worldwide population. Involuntary, rhythmic oscillations centered around one or more joints during postural or kinetic action are the classic phenotypes of ET. Surgical resection or deep brain stimulation (DBS) is often an option for medically refractory ET. This study focuses on the micro-lesion effect, the improvement of pathological symptoms after electrode implantation and before initiation of electrical stimulation. We hypothesize that DBS delivered in response to pathology can ameliorate symptoms and provide additional benefits. Herein, we demonstrate that an external wearable sensor can provide a reliable control signal for responsive DBS. Additionally, we show that with external sensors, micro-lesion effects and underlying tremor differences between the contralateral and ipsilateral implantation side can be observed. **Method** Data was collected from ET subjects during unilateral VIM electrode implantation for DBS at University of Florida Health Shands Hospital. During surgery, external wireless sensors that recorded electromyography (EMG) and acceleration data were placed on the hand dorsum, flexor carpi radialis, extensor carpi ulnaris, and bicep brachii muscles. The subjects were instructed to perform a series of tasks that provoked their tremor, such as reaching for a cup and finger to nose task. Each task lasted 2-3 minutes with periods of rest throughout the duration of the task. The patient was cued when to use their left or right hand and when to rest. Videos were recorded during the tasks and labeled for tremor onsets. Data was collected during DBS off trials. To align the data with the onset of tremor, a trigger was sent and picked up by both the video and the sensors. **Results** The coherence between the hand acceleration and flexor EMG during the reaching for a cup task for three patients provides a mean to distinguish between the ipsilateral and contralateral upper extremity. The peak coherence for the ipsilateral hand implant is at the patient's respective tremor frequency, obtained from the inertial data. The contralateral limb, which saw a reduction of tremor due to the micro-lesion effect, had an expected decrease in the coherence between muscle pairs. These results suggest there is a clear distinction between EMG and acceleration correlates recorded from muscles that are ipsilateral or contralateral to the site of implantation. This suggests that the micro-lesion effect may have a significant impact on the patients' tremor characteristics, therefore altering the thalamocortical connectivity.

Disclosures: S. Cernera: None. **E. Opri:** None. **A. Gunduz:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.15/DP06/Y15 (Dynamic Poster)

Topic: C.04. Movement Disorders

Support: The Grainger Foundation

Title: Calcium imaging of striatal activity evoked by subthalamic nucleus deep brain stimulation

Authors: *J. TREVATHAN[1], E. N. NICOLAI[2], A. J. ASP[4], D. CHENG[5], M. J. SCHACHTER[5], J. J. NASSI[5], S. L. OTTE[6], J. G. PARKER[7], J. LUJAN[3], K. A. LUDWIG[3]
[2]Dept. of Neurologic Surgery, [3]Neurologic Surgery, [1]Mayo Clin., Rochester, MN; [4]Biomed. Engin., Mayo Grad. Sch., Rochester, MN; [5]Inscopix, Inc., Palo Alto, CA; [6]Inscopix, Palo Alto, CA; [7]Dept. of Biol., Stanford Univ., Palo Alto, CA

Abstract: Introduction: Subthalamic nucleus (STN) deep brain stimulation (DBS) is a common surgical treatment for Parkinson's disease that achieves therapeutic benefit through the application of high frequency electrical stimulation. Although, changes in activity from striatal neurons occur following 6-hydroxydopamine (6-OHDA) induced lesioning, a common model of Parkinson's disease, the role of these neurons in the therapeutic response to Parkinson's disease is not well understood. In particular, medium spiny neurons (MSN), the primary striatal outputs, have been shown to form spatially compact coherent clusters encoding locomotion-related information in healthy mice (Barbera et al. 2016). However, in 6-OHDA lesioned animals, MSNs exhibit decreased clustering and specificity (Cho et al. 2002). Here, we demonstrate the first use of calcium imaging to study the effects of DBS on striatal neurons by imaging the stimulation-evoked changes in neural activity with a millisecond resolution.
Methods: Calcium imaging was performed with an nVoke system (Inscopix, Palo Alto, CA) in four 6-OHDA lesioned mice. First, a dopaminergic lesion was performed by injecting 6-OHDA in the substantia nigra pars compacts (SNc). Additionally, dorsal striatal neurons were transfected with GCaMP6m using the adeno-associated virus AAV9. Two weeks after injection, a 1mm gradient index (GRIN) lens was implanted directly above the dorsal striatum and a bipolar stimulation electrode (PlasticsOne, Roanoke, VA) was implanted in the STN. Calcium imaging was performed beginning two weeks after the lens implant. In order to observe stimulation evoked changes in striatal neuron activity recordings were first performed under isoflurane anesthesia and later in behaving animals.
Results: Stimulation evoked neuronal activity was observed in anesthetized animals as a result of STN DBS. Evoked activity showed a non-linear dependency on stimulation parameters.
Conclusion: An animal model combining deep brain optical imaging and DBS was successfully implemented to study changes in neuronal activity occurring at the circuit level. This work represents a first step in studying the effects of stimulation on deep brain activity via optical imaging techniques in freely behaving models of DBS.

Disclosures: J. Trevathan: None. **E.N. Nicolai:** None. **A.J. Asp:** None. **D. Cheng:** A. Employment/Salary (full or part-time):; Inscopix. **M.J. Schachter:** A. Employment/Salary (full or part-time):; Inscopix. **J.J. Nassi:** A. Employment/Salary (full or part-time):; Inscopix. **S.L. Otte:** A. Employment/Salary (full or part-time):; Inscopix. **J.G. Parker:** None. **J. Lujan:** None. **K.A. Ludwig:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.16/Y16

Topic: C.04. Movement Disorders

Title: Precise MRI-based stereotaxic surgery in large animal models

Authors: A. N. GLUD[1], J. BECH[1], L. TVILLING[1], H. ZAER[1], D. ORLOWSKI[1], L. M. FITTING[1], D. ZIEDLER[2], M. GENESER[2], R. SANGILL[2], A. K. O. ALSTRUP[3], C. R. BJARKAM[4], *J. SORENSEN[5]
[1]Dept. of Neurosurg., Aarhus Univ. Hospital, CENSE Group, Aarhus, Denmark, Aarhus C, Denmark; [2]Dept. of Neuroradiology, [3]PET-centre, Aarhus Univ. Hosp., Aarhus, Denmark; [4]Inst. of Clin. Med., Aalborg University, Denmark; [5]Aarhus Univ. Hospital, Head-Neuro Ctr., Aarhus C, Denmark

Abstract: Stereotaxic neurosurgery in large animals is used widely in different sophisticated models, where precision is becoming more crucial as desired anatomical target regions are becoming smaller. Individually calculated coordinates are necessary in large animal models with cortical and subcortical anatomical differences. We present a convenient method to make an MRI-visible skull fiducial for 3D MRI-based stereotaxic procedures in larger experimental animals. Plastic screws were filled with either copper-sulphate solution or MRI-visible paste from a commercially available cranial head marker. The screw fiducials were inserted in the animal skulls and T1 weighted MRI was performed allowing identification of the inserted skull marker.
Results: Both types of fiducial markers were clearly visible on the MRI´s. This allows high precision in the stereotaxic space. The use of skull bone based fiducial markers gives high precision for both targeting and evaluation of stereotaxic systems. There are no metal artifacts and the fiducial is easily removed after surgery.
Conclusion: The fiducial marker can be used as a very precise reference point, either for direct targeting or in evaluation of other stereotaxic systems.
Discussion: Large animal models of brain diseases are in need of reliable methods for precision targeting of deep brain targets. The fiducial marker can work alone or as a control marker in corporation with framebased fiducial markers, or when other systems with possible MRI distortion artifacts are used in the experimental setup.

Disclosures: **A.N. Glud:** None. **J. Bech:** None. **L. Tvilling:** None. **H. Zaer:** None. **D. Orlowski:** None. **L.M. Fitting:** None. **D. Ziedler:** None. **M. Geneser:** None. **R. Sangill:** None. **A.K.O. Alstrup:** None. **C.R. Bjarkam:** None. **J. Sorensen:** A. Employment/Salary (full

or part-time):; CENSE group, Department of Clinical Medicine, Aarhus University, Department of Neurosurgery, Aarhus University Hospital..

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.17/Y17

Topic: C.04. Movement Disorders

Support: NIH BRAIN Initiative UH3NS095553

Title: Towards an adaptive cortico-thalamic closed-loop deep brain stimulation for the treatment of essential tremor

Authors: *E. OPRI[1], S. L. CERNERA[1], M. OKUN[1], K. FOOTE[1], A. GUNDUZ[2]
[2]Biomed. Engin., [1]Univ. of Florida, Gainesville, FL

Abstract: Essential tremor (ET) is defined as a rhythmical, involuntary oscillatory movement of the limbs and is one of the most common movement disorders. Intention tremor occurs mostly in the upper limbs (with slow oscillations between ~4-12 Hz) during the initiation and execution of goal-directed reaching motions, while it is absent at rest. It has currently been suggested that a synchronous pathological oscillation in a network that includes the premotor (PM) and primary motor (M1) cortices, the ventral intermediate nucleus (Vim), and the cerebellum is suppressed through deep brain stimulation (DBS) by jamming the “tremor cells” in the thalamus. Eight ET patients underwent DBS implantation surgery with the addition of ipsilateral cortical strip placement. A subgroup of patients was also implanted with Activa PC+S neurostimulator. Hence, we show the feasibility of a closed-loop system using thalamocortical neuromarkers evoked during different behavioral tasks, such as moving a hand or reaching a cup, to enable the control of stimulation activation and deactivation. The closed-loop system is being developed to run fully onboard the Activa PC+S device. In addition, analysis between PM, M1 and Vim shed light on the relationship and network between the regions involved in the tremor generation. As a failsafe solution, we are also exploring the use of additional external sources of input for the detection, such as inertial sensors. The fusion between internal and external inputs allows the implementation of “context aware” stimulation patterns, also useful in improving the quality of life of the patients. Our results suggest that the approach proposed would lead to prolonged battery life, fewer stimulation side effects (e.g. balance and speech impairment), while delivering an equally effective treatment.

Disclosures: E. Opri: None. **S.L. Cernera:** None. **M. Okun:** None. **K. Foote:** None. **A. Gunduz:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.18/Y18

Topic: C.04. Movement Disorders

Support: NIH R01 NS096008

NSF CAREER 1553482

Title: Closed-loop deep brain stimulation paradigm for Tourette syndrome

Authors: *J. CAGLE, M. S. OKUN, K. D. FOOTE, A. GUNDUZ
Univ. of Florida, Gainesville, FL

Abstract: Tourette syndrome (TS) is a continuous lifelong condition that is highly prevalent and socially embarrassing. Although in most cases motor and vocal tics wane by the late teenage year, some TS sufferers with motor and vocal tics are resistant to medication and to behavioral intervention and the issues persist into adulthood. Deep brain stimulation (DBS) has emerged as a promising treatment option for addressing tics in appropriately screened cases. The neurostimulator, Medtronic Activa PC+S, is a highly novel next generation technology, as it can record brain activity and can be programmed to provide stimulation in response to pathological activity (i.e., responsive stimulation) instead of the standard 24-hour continuous stimulation. Therefore, we proposed the responsive stimulation paradigm for TS using a novel, bidirectional neural interface, which can record LFPs, while simultaneously delivering DBS therapy.Three adult TS patients were recruited for the study. Both deep brain leads and cortical ECoG strips were implanted as part of the responsive stimulation system. LFPs that are time synchronized with tic behavior were collected from the centromedian thalamic (CM) region using DBS leads, and from the primary motor (M1) cortex using chronically implanted ECoG strips. Simultaneous recordings of electromyogram (EMG), accelerometer (ACC), videos, and LFP were collected from adult TS patients recruited in the study. All recordings were time-synchronized for better identification of tic features, and all videos are labeled by the clinicians. Power spectral density prior to tic onset were computed every 500 milliseconds with 250 milliseconds increment, and the frequency band with highest correlation to tic onset were chosen as tic features for detection. A human tic detector were built with the features identified and implemented in Activa PC+S, and the tic severity scores were rated by blinded psychiatrist for both traditional continuous stimulation and the novel responsive stimulation paradigms.The 20 features within 3 seconds prior to Tic onset with highest correlation to the presence of tic includes Beta (20-25Hz) and Alpha (8-12Hz). Beta band shows most promising results as it captures the Tic better than other frequency bands. The tic severity scores for the responsive stimulation paradigm using cortical beta power as detection appears to be no worse than current continuous stimulation paradigm.

Therefore, we conclude that responsive stimulation is as effective as traditional continuous stimulation paradigm, and it can prolong battery-life of the neurostimulator and reduce side-effects on the patients.

Disclosures: **J. Cagle:** None. **M.S. Okun:** None. **K.D. Foote:** None. **A. Gunduz:** None.

Poster

389. Movement Disorders II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 389.19/Z1

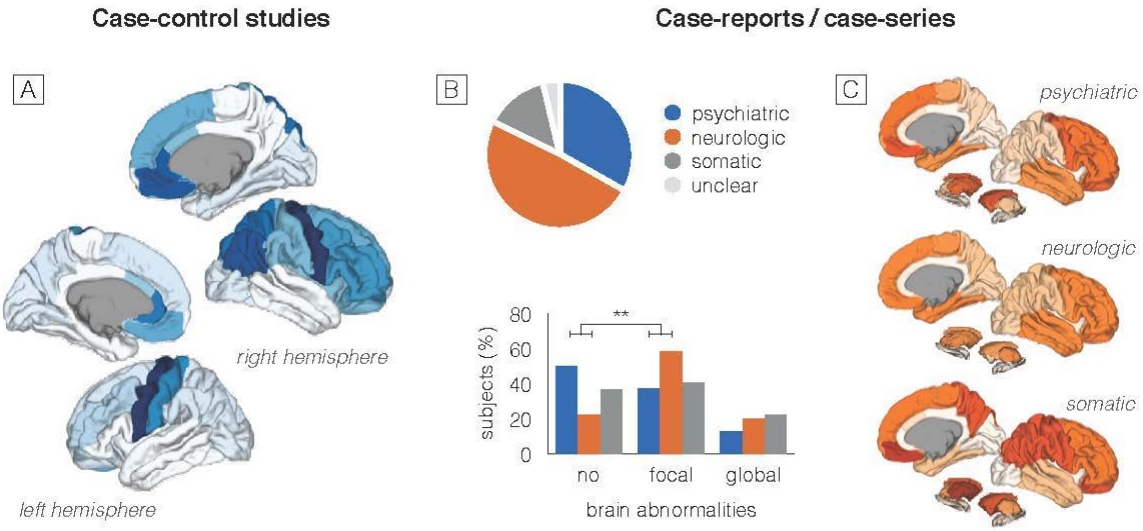
Topic: C.04. Movement Disorders

Title: Brain abnormalities in catatonia across psychiatric, neurological, and general medical conditions: A systematic review and meta-analysis of neuroimaging findings

Authors: X. CHANG[1], *G. COLLIN[2]
[1]Univ. Med. Ctr. Utrecht, Utrecht, Netherlands; [2]BIDMC, Harvard Med. School, Dept. of Psychiatry, Boston, MA

Abstract: Objective: Catatonia is a striking psychomotor syndrome characterized by dysregulation of motor and behavioral functions. The syndrome occurs in psychiatric, neurologic, and general medical conditions, with marked similarities in symptoms and treatment response, suggesting a common neurobiological substrate. This review explores brain abnormalities in catatonia across clinical conditions.
Methods: A systemic literature search was performed in Pubmed and Embase databases. Relevant documentation published in English up to July 2016 was extracted. A selection procedure was performed to identify relevant articles reporting on brain imaging in catatonia patients. Neuroimaging results were assessed across diagnostic groups and structural vs. functional imaging methods.
Results: A total of 8 case-control studies and 179 case-reports/series were identified. The case-control studies demonstrated abnormalities in sensorimotor, prefrontal, and parietal cortex (more pronounced in right hemisphere) in a total of 58 catatonia patients, as compared to 96 healthy controls and 50 non-catatonic psychiatric controls (Fig. 1A). The case-reports/series describing 197 individual cases with a variety of conditions, showed global or focal brain abnormalities in around two-thirds of cases (Fig. 1B). There was an association between diagnostic group and imaging results ($\chi^2 = 14.25$, $p = .007$), such that the proportion of focal abnormalities was higher in neurologic than psychiatric cases ($\chi^2 = 10.53$, $p = .001$). The regional distribution of abnormalities in frontal, parietal, and temporal cortex, and basal ganglia, was similar across groups (Fig. 1C).
Conclusions: This review shows that global and focal brain abnormalities are common in catatonia across clinical conditions. Neuroimaging results implicate distributed regions in mainly

frontal and parietal cortex, with more pronounced alterations in right hemisphere. The distribution of focal abnormalities was similar for diagnostic groups, consistent with a common neurobiological substrate across clinical conditions.



Disclosures: **X. Chang:** None. **G. Collin:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.01/Z2

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: JSPS KAKENHI JP16K08635

JSPS KAKENHI JP17K07123

JSPS KAKENHI JP16K11308

JSPS KAKENHI JP15H04999

Takeda Science Foundation

Title: Role of glial and neuronal TrkB signaling in protection of retinal ganglion cells following optic nerve injury

Authors: *Y. AZUCHI[1,2], A. KIMURA[1], C. HARADA[1], K. NAMEKATA[1], T. HARADA[1]
[1]Visual Res. Project, Tokyo Metropolitan Inst. of Med. Sci., Tokyo, Japan; [2]Envrn. Sci., Grad. Sch. of Science, Toho Univ., Chiba, Japan

Abstract: Glaucoma is characterized by progressive degeneration of retinal ganglion cells (RGCs) and their axons. The optic nerve injury (ONI) model mimics some aspects of glaucoma, including RGC death induced by excitotoxicity and oxidative stress, and therefore, it is a useful animal model for glaucoma. Previous studies have shown that trophic factors, such as brain-derived neurotrophic factor (BDNF) protect RGCs and promote axon regeneration in an ONI model. BDNF is known to regulate neural cell survival and axonal outgrowth mainly by activating TrkB receptors. However, the detailed roles of TrkB in different cell types during retinal diseases such as traumatic injury and glaucoma are still unknown. To investigate the TrkB function in neurons and glial cells, we used two conditional knockout mouse lines, in which TrkB was deleted from retinal glia ($TrkB^{GFAP}$ KO) or retinal neurons ($TrkB^{c\text{-}kit}$ KO). Deletion of TrkB specifically from neurons including RGCs shows no developmental abnormalities and similarly, mice with selective TrkB deletion from glia including Müller glia are viable. We then examined the neuroprotective effect in these mouse lines using an ONI model. Histochemical analysis revealed that compared with WT mice at 7 days after ONI, more severe RGC loss was detected in both mouse lines with the similar levels. These findings suggest that not only neuronal TrkB but glial TrkB also has critical roles for RGC protection during ONI. However, we found that while loss of RGCs continued to increase with time in $TrkB^{c\text{-}kit}$ KO mice compared with WT, no further loss was observed in $TrkB^{GFAP}$ KO mice at 14 days after ONI. Next, we examined if the lack of TrkB signaling in retinal glia or retinal neurons affect expression of retinal neurotrophic factors following ONI. In WT mice, the expression of basic fibroblast growth factor (bFGF) is upregulated at 3 days after ONI. We previously reported that the increase in bFGF is suppressed in $TrkB^{GFAP}$ KO mice, indicating that the main source of bFGF following ONI is glia and it is mediated via glial TrkB signaling. On the other hand, there was no difference in this upregulation between $TrkB^{c\text{-}kit}$ KO and WT mice, suggesting that elevation of bFGF is independent of the neuronal TrkB signaling. Our findings provide the possibility that glial and neuronal TrkB have different pathways on RGC protection and operate at different time points.

Disclosures: Y. Azuchi: None. **A. Kimura:** None. **C. Harada:** None. **K. Namekata:** None. **T. Harada:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.02/Z3

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: CNPq/INCT/INPeTAm, Grant 573695/2008-3

FAPERGS - PRONEX/FAPERGS/CNPq, Grant 10/0044-3

Title: N9 murine microglial cell activation is attenuated by rosmarinic acid through down regulation of inflammatory cytokines and cleaved caspase-3

Authors: *P. PEREIRA[1], V. R. COELHO[1], C. M. VIAU[1], R. B. STAUB[2], M. S. DE SOUZA[2], P. F. PFLÜGER[1], J. SAFFI[2]
[1]Federal Univ. of Rio Grande Do Sul, Porto Alegre, Brazil; [2]Federal Univ. of Hlth. Sci. of Porto Alegre, Porto Alegre, Brazil

Abstract: Rosmarinic acid (RA) has shown several biological activities, including antioxidant and anti-inflammatory properties and protective effects in many types of tissue like the brain tissue. Microglial activation has been considered a crucial process in the pathogenesis of neuroinflammation and psychiatric disorders. Microglia could be activated into the classic activated cytotoxic state M1 or the alternative activated neurotrophic state M2. The present study evaluated the ability of RA to inhibit the microglia activation induced by lipopolysaccharide (LPS) in the N9 murine microglial cell line and investigated possible mechanisms involved in this process. The cell line was obtained by immortalization of embryonic brain cultures with 3RV retrovirus carrying an activated v-myc oncogene and was grown in RPMI. Cells (1×10^4 cells/mL) were seeded on 6-well tissue culture plates and grown for 1 day up to 70-80% confluence before treatment with RA. In all tests, N9 murine microglial cells were pretreated with RA (0.1, 1.0, and 10 μM) for 20 hours and exposed to LPS (1μM/mL) for 4 h. Cell viability was measured by Trypan blue exclusion assay (TB). Reactive oxygen species (ROS) detection, quantification of cleaved caspase-3, and mitochondrial electrochemical potential were performed by flow cytometric analysis. iNOS, Arg-1, TNF-α, IL1-β, and IL-6 proteins were examined by Western blotting analysis and their antigens were detected using the chemiluminescence technique. Image analysis was performed using GraphPad Prism v5 program. The effect of RA on DNA was evaluated by Comet assay. In this study, RA attenuated the expression of the M1 marker iNOS and the levels of pro-inflammatory factors including TNF-α, IL1-β, and IL-6; it increased the expression of the M2 marker Arg-1, and inhibited, at least in part, ROS generation and loss of mitochondrial outer membrane permeabilization (MOMP) through the inhibition of cleaved caspase-3 activation. RA also inhibited DNA damage reassuring cell protection. Thus, the results suggested a protective effect of RA down regulating inflammatory cytokines and cleaved caspase-3.

Disclosures: **P. Pereira:** None. **V.R. Coelho:** None. **C.M. Viau:** None. **R.B. Staub:** None. **M.S. de Souza:** None. **P.F. Pflüger:** None. **J. Saffi:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.03/Z4

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: MOST1012320B010041MY3

V106D21-002-MY3-1

Title: FKBP51 mediates neurotoxicity, astrogliosis, and demyelination via cell type-dependent signaling pathway in excitotoxicity-induced neurodegeneration

Authors: *S.-H. LIN[1], H.-Y. CHUANG[1], Y.-M. KUO[1,5], P.-C. HSU[1,2], I.-H. LEE[6], A.-Y. LIN[3], C.-J. JENG[4], Y.-H. LEE[1,2]
[2]Brain Res. Ctr., [3]Pharmacol., [4]Cell Biol. and Anat., [1]Natl. Yang-Ming Univ., Taipei, Taiwan; [5]Anesthesiol., [6]Neurol., Taipei Veteran Gen. Hosp., Taipei, Taiwan

Abstract: Excitotoxicity induced by glutamate overflow is the most common cause of neurodegeneration following both acute and chronic brain injury. FK506 binding protein 51 (FKBP51), encoded by *Fkbp5* gene, is a cochaperone best known for regulating glucocorticoid receptor (GR) and Akt-dephosphorylating phosphatase. Genetic variants of *Fkbp5* causing FKBP51 elevation have been found in patients with neuropsychiatric disorders, and yet its role in the excitotoxicity-induced neurodegeneration remained unknown. We used intracerebroventricular injection of kainic acid (icv-KA)-induced epilepsy mouse model on wild type (WT) and *Fkbp5*-knockout (*Fkbp5*-KO) mice to investigate the role of FKBP51 in excitotoxicity-induced neurodegeneration. Our data show that *Fkbp5*-KO mice are highly resistant to KA-induced hippocampal damage and demyelination in the white matter regions (corpus callosum and cingulum), accompanied with much weaker astrogliosis in the hippocampus and attenuated seizure activity. Similar injury-resistant effect of *Fkbp5*-KO was observed in the transient hypoxia-ischemia (tHI)-induced brain injury mouse model with much improved survival rate and lessened cerebral infarction. These neuroprotective effects of *Fkbp5*-KO were reproduced in primary mouse cortical neurons, neuron-glia mix culture, and myelin-forming oligodendrocytes (OLs) treated with excitotoxic NMDA and AMPA, respectively. Mechanistic studies further revealed that icv-KA induced GR reduction and Akt phosphorylation in the corpus callosum and hippocampus in a FKBP51-dependent manner, respectively. The FKBP51-dependent GR reduction in the corpus callosm was reproduced in vitro in AMPA-treated OLs, in which FKBP5KO showed elevation of GR signaling and downstream prosurvival gene expression to attenuate OL damage. On the other hand, the FKBP51-dependent Akt phosphorylation effect was found in the NMDA-treated astrocyte-neuron mix culture, but not neuron-enriched culture, accompanied with the NFkB activation and astrocyte reactivation. In

conclusion, these data suggest that *Fkbp5*/FKBP51 mediates excitotoxicity via cell type-dependent regulations of GR and Akt signaling and glia-neuron interaction. Genetic variations of *FKBP5* gene may contribute to the individual susceptibility to insult-induced neurodegeneration.

Disclosures: S. Lin: None. **H. Chuang:** None. **Y. Kuo:** None. **P. Hsu:** None. **I. Lee:** None. **A. Lin:** None. **C. Jeng:** None. **Y. Lee:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.04/Z5

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: Foundation Illinois Society for the Prevention of Blindness

Dr. John P. and Therese E. Mulcahy Endowed Professor in Ophthalmology

Richard A. Peritt Charitable Foundation

Title: Intracellular signaling in astrocytes in response to elevated hydrostatic pressure

Authors: *S. KAJA[1,3,2], V. R. RAO[2,3,1], Y. NAUMCHUK[2], A. J. PAYNE[4]
[1]Ophthalmology, [2]Mol. Pharmacol. & Therapeut., Loyola Univ. Chicago, Maywood, IL; [3]Res. Service, Edward Hines Jr. VA Hosp., Hines, IL; [4]K&P Scientific LLC, Oak Park, IL

Abstract: Primary open angle glaucoma (POAG) is associated with elevated intraocular pressure (IOP), manifesting in a pathological triad of optic nerve head remodeling, damage to the optic nerve, and retinal ganglion cell (RGC) loss. Optic nerve head astrocytes (ONHAs), the primary cell type in the optic nerve head, undergo significant pathological changes in POAG. Here, the cellular and molecular consequences of elevated hydrostatic pressure on cultured ONHAs were investigated. To this end, primary adult rat ONHAs were exposed to ambient or elevated hydrostatic pressure (25-30 mm Hg above ambient pressure) for 2 – 30 hr using a cell culture pressure chamber. Cell viability and proliferation were quantified using MTT and lactate dehydrogenase (LDH) release assays, while levels of oxidative stress and nitric oxide (NO) were quantified using fluorescent sensors, CellROX®, H_2DCFDA and DAF-FM.
ONHA cultures exposed to elevated hydrostatic pressure for up to 30 hr did not exhibit altered cell viability, and LDH release was similar between cells exposed to ambient or elevated hydrostatic pressure ($P = 0.651$). However, elevated hydrostatic pressure significantly increased the sensitivity to a subsequent oxidative challenge in the MTT assay ($P < 0.01$) and the LDH assay ($P < 0.01$). Subsequent analysis of ROS levels revealed that elevated hydrostatic pressure caused a statistically significant increase in the level of ROS as early as 2 hr after exposure to elevated hydrostatic pressure, as quantified by CellROX® fluorescent staining and DCFDA

fluorescence. Furthermore, hydrostatic pressure resulted in an up-regulation of nitric oxide synthase 2 (NOS2) with a concomitant increase in NO levels, as assessed by DAF-FM. Our data clearly show that elevated hydrostatic pressure increases the levels of ROS and NO in cultured ONHAs *in vitro*. These data suggest that even modest exposure to elevated IOP in POAG may significantly alter the oxidation response of ONHAs and accelerate neurotoxic signaling early during glaucoma pathogenesis.

Disclosures: **S. Kaja:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); K&P Scientific LLC, Experimentica Ltd.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); K&P Scientific LLC, Experimentica Ltd.. F. Consulting Fees (e.g., advisory boards); Experimentica Ltd.. **V.R. Rao:** None. **Y. Naumchuk:** None. **A.J. Payne:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); K&P Scientific LLC.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.05/Z6

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH AG 022550

NIH AG 027956

Title: Connexin 43 as a mediator of androgen and estrogen-induced protection against oxidative stress in astrocytes

Authors: *N. K. KUBELKA, N. RYBALCHENKO, M. SINGH
Ctr. for Neurosci. Discovery, Univ. of North Texas Hlth. Sci. Ctr., Fort Worth, TX

Abstract: Connexin 43 (Cx43) is a transmembrane protein that is a component of gap junctions and highly expressed in astrocytes. While evidence exists supporting Cx43 in the regulation of non-central nervous system (CNS) cell viability, the roles of Cx43 in CNS protection are not well understood. Our laboratory has described that both estrogens and androgens have protective effects in astrocytes, and that this protection is afforded, in part, by reducing oxidative load. Given the abundance of Cx43, and its role regulating oxidative burden, we determined if the protective effects of either dihydrotestosterone (DHT), its metabolite, 3-beta diol, or estradiol (E2) in astrocytes are mediated by Cx43. Using cortical astrocytes derived from C57Bl/6 mice, we first assessed the effects of these three hormones on the expression of Cx43 mRNA. RTPCR analysis revealed that 3 hr treatment with E2 (100nM) resulted in a significant increase in Cx43

mRNA expression relative to control, while DHT and 3βdiol did not. In order to determine if Cx43 was a key mediator of the protective effects of either hormone, we used conformation-preferring inhibitors of Cx43. While neither E2 alone (100nM) nor Gap19 alone (hemichannel selective Cx43 inhibitor peptide, 10μM) protected against IAA toxicity, application of both E2 and Gap19 resulted in significant protection against IAA toxicity, suggesting a facilitation of E2's protective effects through inhibition of Cx43 hemichannels. The effects of DHT were similarly enhanced in the presence of the Cx43 hemichannel inhibitor (Gap19). When the conformation non-selective inhibitor of Cx43, Gap26, (which inhibits both Cx43 hemichannels and gap junctions) was used, Gap26 alone resulted in significant protection, and protection was further enhanced in astrocytes treated with both Gap26 and E2. As a surrogate measure of Cx43 function, we also evaluated if the three hormones elicited changes in the phosphorylation state of Cx43 using a Cx43 Ser368 phosphospecific antibody. Our data describe, for the first time, the involvement of Cx43 in the cytoprotective effects of E2 and DHT in the brain. As such, regulation of Cx43 may be a mechanistic component of hormone-induced brain protection, and regulating its function may be a crucial component of hormone therapy.

Disclosures: **N.K. Kubelka:** None. **N. Rybalchenko:** None. **M. Singh:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.06/Z7

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: CONACyT-FOSSIS 262010

Title: 3-hydroxykynurenine and 3-hydroxyanthranilic acid enhance the toxicity induced by copper in rat astrocytes culture

Authors: *D. RAMÍREZ ORTEGA[1], A. SALAZAR RAMIRO[2], D. GONZALEZ ESQUIVEL[3], C. RIOS[3], B. PINEDA[2], V. PEREZ DE LA CRUZ[3]
[1]Inst. Nacional De Neurología y Neurocirugía Manuel Velasco, México, Distrito Federal, Mexico; [2]NEUROINMUNOLOGIA, [3]NEUROQUIMICA, INSTITUTO NACIONAL DE NEUROLOGIA, MEXICO CITY, Mexico

Abstract: Copper is a heavy metal and an integral component of various enzymes. It is necessary for mitochondrial respiration and other biological functions; however, excess copper is neurotoxic and has been implicated with neurodegenerative diseases as Alzheimer. This metal is able to modify the cellular redox environment, which can influence various cellular functions, signaling and catabolic pathways. In this context, tryptophan degradation through kynurenine pathway (KP) is modulated by the redox environment and produces some metabolites with redox

properties as 3-hydroxykynurenine (3-HK) and 3-hydroxyantrhanilic acid (3-HANA). The imbalance in the production of these kynurenines is related with some neuropathologies, in which the common factors are oxidative stress, inflammation and cell death. The aim of this study was to evaluate the effect of these kynurenines on the cupper toxicity in astrocytes cultures. First, we evaluated the $CuSO_4$ (0-500 µM) effect on MTT reduction, ROS production, mitochondrial membrane potential (MMP) and cell viability on primary cultured astrocytes. Then was evaluated the effect of the co-incubation of $CuSO_4$ (350 µM) whit 3-HK and 3-HANA (100 µM) in the same parameters that were previously tested and also GHS levels. In addition, the chelating copper effect of 3-HK and 3-HANA was evaluated. Our results showed that $CuSO_4$ decreased MTT reduction and MMP, while it increased ROS production and cell death in a concentration-dependent manner. The co-incubation with 3-HK and 3-HANA enhances the toxic effect of copper in MTT reduction, MMP and cell death. Copper also decreased GSH levels around 50%, but the co-incubation with the kynurenines decreased 70% GSH levels. However, the increase in ROS production induced by copper was abolished by 3-HK and 3-HANA. Both 3-HK and 3-HANA are able to chelate cupper in a concentration dependent manner. These data suggest that 3-HK and 3-HANA increased copper toxicity in an independent manner to ROS production; however, their effect on GSH levels could play an important role in the potentiation of cell damage induced by this metal.

Disclosures: D. Ramírez Ortega: None. **A. Salazar Ramiro:** None. **D. Gonzalez Esquivel:** None. **C. Rios:** None. **B. Pineda:** None. **V. Perez de la Cruz:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.07/Z8

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: USAMRMC Project Grant (Contract#W81XWH-07-2-0078

Title: Co-culturing with Schwann cells confers protection on dorsal root ganglion neurons against cytotoxicity induced by silver nanoparticles

Authors: *J. C. LAI[1], W. GAO[2], S. W. LEUNG[2]
[1]Idaho State Univ. Col. of Pharm., Pocatello, ID; [2]Civil & Envrn. Engin., Idaho State Univ. Col. of Sci. & Engin., Pocatello, ID

Abstract: Our previous studies have demonstrated that various nanomaterials, including metallic nanoparticles, can induce cytotoxicity in different neural cell types although their underlying neurotoxic and molecular mechanisms are far from being understood. In this study, we developed a co-culture model consisting of immortalized dorsal root ganglion (DRG) neurons

and Schwann cells and employed it to investigate our hypothesis that co-culturing DRG neurons with Schwann cells confers protection on these neurons against neurotoxicity induced by silver nanoparticles. Our results suggested when exposed to silver nanoparticles, cell survival was enhanced in co-culture compared to that in monotypic cultures. Synapsin I expression was increased in DRG neurons when they were co-cultured with Schwann cells and treated with or without nanoparticles. Glial fibrillary acidic protein (GFAP) expression was increased in Schwann cells when they were co-cultured with DRG neurons and treated with nanoparticles. The expression of ERK and p-ERK was altered in treated DRG neurons, Schwann cells, and DRG neurons co-cultured with Schwann cells. Co-culturing with Schwann cells induced morphological differentiation in DRG neurons and rendered them less susceptible to morphological changes and damage induced by silver nanoparticles. Thus, consistent with our hypothesis, our results may assume pathophysiological significance in peripheral nerve degeneration and neurodegeneration induced by silver nanoparticles.

Disclosures: J.C. Lai: None. **W. Gao:** None. **S.W. Leung:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.08/Z9

Topic: C.01. Brain Wellness and Aging

Support: 2R01AG033649-07

Title: Repeated daily doses of intranasal insulin aspart in young and aged F344

Authors: *K. L. ANDERSON, H. N. FRAZIER, A. O. GHOWERI, K. E. HARGIS, J. C. GANT, L. D. BREWER, N. M. PORTER, E. M. BLALOCK, O. THIBAULT
Pharmacol. and Nutritional Sci., Univ. of Kentucky, Lexington, KY

Abstract: In an attempt to combat decreased insulin signaling in the brain of Alzheimer's disease (AD) patients, several groups have used intranasal insulin in clinical and pre-clinical settings. Results show improved memory in young volunteers, patients with mild cognitive impairment, as well as in animal models of aging, diabetes or AD. We have previously reported that intranasal delivery of different insulin formulations (short and long acting), under either acute (single dose) or short term (9 doses) conditions, can enhance insulin signaling in the brain, alter cerebral blood flow and enhance memory recall. Here, we tested whether long term intranasal insulin (>60 doses) provided a greater enhancement in memory and recall in young and aged animals, or if the continued presence of the ligand might cause receptor downregulation or desensitization. Further, it is not clear how intranasal insulin alters functional communication, improves learning, or facilitates memory retrieval, particularly in animal model of aging. Our

study was therefore designed to address these limitations.
We characterized the impact of long-term (3 months, > 60 daily doses) intranasal insulin aspart (NovoLog®) on behavior using the Morris water maze (MWM) as well as on insulin receptor levels using autoradiography and immunohistochemistry. We also investigated the role of long term daily doses of insulin on hippocampal RNA species using microarray analyses. Saline (0.9%) or insulin aspart (equivalent to 10 IU/day) delivery was accomplished on non-anesthetized animals as previously published by our group (10 uL total volume per day). Learning and memory were evaluated using the classical MWM, and the extended version by including a reversal learning and reversal probe session in young and aged F344 (n=23/age). MWM training started on the 59th dose and study was terminated after the 62-64th dose. As previously shown, young (5 months) and aged (21 months) rats showed differences in learning and memory. Long term intranasal insulin did not raise peripheral blood glucose levels but did ameliorate aspects of memory recall in the aged animals.
These studies address the impact of repeated daily doses across 3 months in young and aged animals and directly test whether measurable changes in insulin signaling/sensitivity/binding occur across age and under conditions which should normally give rise to receptor desensitization. The results further highlight differences in insulin signaling between the brain and the periphery. We suggest that chronic changes in brain insulin using intranasal delivery can inform on therapies designed to increase insulin signaling and redress cognitive decline with aging.

Disclosures: K.L. Anderson: None. **H.N. Frazier:** None. **A.O. Ghoweri:** None. **K.E. Hargis:** None. **J.C. Gant:** None. **L.D. Brewer:** None. **N.M. Porter:** None. **E.M. Blalock:** None. **O. Thibault:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.09/Z10

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant R01AG004542

NIH Grant 5R01AG033649

Title: *In vivo* neuronal and astrocytic high-resolution calcium in aging and a model of AD

Authors: *J. C. GANT[1], P. SOMPOL[2], P. W. LANDFIELD[3], C. M. NORRIS[4], O. THIBAULT[5]
[1]Molec. and Biomed. Pharm, [3]Pharmacol. and Nutritional Sci., [2]Univ. of Kentucky, Lexington,

KY; [4]Sanders-Brown Ctr. Aging, Univ. Kentucky, Lexington, KY; [5]Dept. of Pharmacol. and Nutritional Sci., Univ. Kentucky Med. Ctr., Lexington, KY

Abstract: Measuring fluorescence using traditional epifluorescence microscopes and charge-coupled devices has long been the standard for quantifying ion changes in neurons and astrocytes during bouts of activation. This technique is well aligned with electrophysiological approaches such that the ion indicator can be delivered to a single cell during the recording process. Even when combined with confocal microscopy, and increased signal to noise, however, this approach lacks in temporal resolution needed to lock imaging with individual action potentials at physiologically relevant frequencies (3-50 Hz). Perhaps more importantly, this approach limits recording and imaging to a single cell. However, the field of Neuroscience has been stepping away from single cell recording and imaging, and has moved toward collection of data from multiple cells and cell ensembles, both *in vivo* and *ex vivo* using multiphoton microscopy. In order to provide higher resolution data on the relationship between cells that are activated in the brain and form the neurovascular unit, with assistance from Scientifica we performed experiments on young and aged rats as well as mice (2x transgenic APP/PS1 mice) both ex vivo and in vivo. We investigated the relationship between neurons and astrocytes, neurons and blood vessels, astrocytes and blood vessels. Because the temporal and spatial relationships between these arrays are best characterized during periods of activity with stimulated some of the tissues at different frequencies (3-50 Hz). Following stereotaxic delivery of GCaMP6s (neurons) and GCaMP6f (astrocytes) 4-6 weeks prior to imaging, and in combination with acute tail vein delivery of rhodamine dextran red, we conducted frequency tuning curves in hippocampal slices from young and aged animals, and examined the relationship between blood vessels and astrocytes in vivo. We describe our results and analysis strategy and also report on the methods used to image calcium changes in young and aged animals, as well as calcium changes in somatosensory cortex of the mice (neurons and astrocytes). For the *ex vivo* work, we focus our analysis on basilar and apical dendrites during activation. The frequency tuning curves for these areas are reported for young and aged animals. For the *in vivo* work in mice, we were able to record spontaneous events in astrocytes and examine the relationship of the astrocytes to the vascular system. These preliminary results demonstrate the feasibility of this approach for studies of aging and AD.

Disclosures: J.C. Gant: None. **P. Sompol:** None. **P.W. Landfield:** None. **C.M. Norris:** None. **O. Thibault:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.10/Z11

Topic: C.01. Brain Wellness and Aging

Support: 2R01AG033649-07

T32-DK007778

P30GM110787

Title: Signaling and expression of a truncated, constitutively active human insulin receptor in hippocampal neurons

Authors: *H. N. FRAZIER[1], K. L. ANDERSON[1], S. MAIMAITI[1], A. O. GOWHERI[1], S. D. KRANER[2], G. J. POPA[3], M. D. MENDENHALL[3], C. M. NORRIS[2], O. THIBAULT[1]
[1]Pharmacol., [2]Sanders Brown Ctr. on Aging, [3]Mol. and Cell. Biochem., Univ. of Kentucky, Lexington, KY

Abstract: Insulin signaling is indispensable in the periphery and it is becoming clear that insulin is also important for normal brain function. Early stage clinical trials report a positive impact of intranasal insulin on memory recall in young subjects and patients with mild cognitive decline or Alzheimer's disease (AD). To address alternative strategies for enhancing insulin signaling in the brain, we have conducted a series of experiments using a constitutively active human insulin receptor (IR). Prior to in vivo experimentation with AAV delivery, lentiviral clones were derived and used to evaluate functional characteristics in rat primary mixed hippocampal cultures.
Cells were infected with either a mammalian expression plasmid encoding a red fluorescence protein (ires-dTomato), or a construct containing the truncated human IR beta subunit (HA-IRβ-ires-dTomato) via a targeted lentiviral system. A synapsin promoter was included to limit expression to neurons. Immunocytochemistry assays using antibody against HA-tagged IRβ confirmed expression of the lentiviral plasmid to neurons. The expression level and effect of IRβ on insulin signaling was confirmed by performing immunocytochemistry and Western immunoblots measuring pAkt/Akt ratio. Whole cell calcium currents were recorded in cultures infected with lentivirus using patch-clamp techniques. Different recording protocols were used to test for specificity of the effect to a subtype of voltage-gated calcium current. To more thoroughly characterize associations between calcium homeostasis and the IR, calcium imaging experiments using Fura-2 were performed. Other outcome measures included direct measures of glucose utilization rates using 2-NDBG.
Lentiviral infection of mixed primary hippocampal cultures was successful for all our constructs. Western blots of infected mixed hippocampal cultures provide evidence that transfection with the truncated IRβ plasmid confers elevated IR signaling compared to controls.
Immunocytochemistry shows the presence of the HA tag in nearly 80% of infected cells. Constitutive activity was also detected. Patch-clamp recordings show calcium currents are a target of insulin receptor activity in IRβ-expressing hippocampal neurons. Calcium levels at rest and during synaptic activity were not altered between the two groups indicating little impact of insulin signaling on resting conditions. Glucose utilization was altered with expression of the constitutively active IR. This characterization provides insights into future intervention approaches to combat cognitive decline in AD and/or aging using molecular methods to enhance insulin signaling.

Disclosures: **H.N. Frazier:** None. **K.L. Anderson:** None. **S. Maimaiti:** None. **A.O. Gowheri:** None. **S.D. Kraner:** None. **G.J. Popa:** None. **M.D. Mendenhall:** None. **C.M. Norris:** None. **O. Thibault:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.11/Z12

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Title: Macroglial Uncoupling Protein 2 deletion increases retinal ganglion cell survival in glaucoma

Authors: *D. HASS, C. J. BARNSTABLE
Neural and Behavioral Sci., The Penn State Col. of Med., Hershey, PA

Abstract: *Introduction*: Glaucoma is a leading cause of irreversible blindness worldwide. Vision loss in glaucoma is associated with elevated intra-ocular pressure (IOP) that leads to retinal ganglion cell (RGC) death. Growing evidence suggests that elevated IOP causes an imbalance in the retinal oxidation state, favoring the production of reactive oxygen species (ROS). ROS may induce toxicity either directly within RGCs, or through Müller glia and astrocytes (macroglia) that normally provide anti-oxidative support to RGCs. Our goal is to assess whether macroglial anti-oxidative support is relevant to the preservation of vision in glaucoma. Uncoupling protein 2 (UCP2) is a mitochondrial antioxidant that regulates ROS production by decreasing the proton motive force. We tested the hypotheses that macroglial UCP2 protects RGCs from oxidative stress and cell death during glaucoma.
Materials and Methods: In the following studies, we used $UCP2^{fl/fl}$; GFAP-$creER^{T2}$ mice injected with tamoxifen or vehicle. Using MitoTracker and $SeahorseXF^{96}$ assays, we identified the mitochondrial phenotype of astrocytes cultured from these mice, normally and when stressed with 5 μM tert-butyl hydroperoxide (tBHP). *In vivo*, we modeled glaucoma by injecting microspheres in to the anterior chamber of the mouse eye to artificially increase IOP. We then measured optomotor responses and the density of RBPMS+ RGCs. To determine whether UCP2 alters reactive gliosis or retinal oxidative stress, we measured GFAP and 4-hydroxynoneol immunoreactivity.
Results: *In vitro*, UCP2 recombination did not alter mitochondrial number, membrane potential, or H^+ leak. tBHP exposure increased H^+ leak and mitochondrial number in a UCP2-dependent manner. *In vivo*, IOP elevation significantly decreased optomotor responses and RGC survival in bead-injected eyes, relative to PBS-injected control eyes. Macroglial UCP2 deficiency was neuroprotective and blunted glaucomatous RGC loss. Additionally, UCP2-deficiency decreased in retinal oxidative stress and reactive gliosis.

Discussion: The *in vitro* results demonstrate that UCP2 uncouples stressed mitochondria, and may increase mitochondrial fission. While this suggests a well-supported protective function for UCP2, our *in vivo* data indicate that macroglial UCP2 impairs RGC survival and other molecular measures of retinal health. Together, these results suggest that glaucomatous RGCs require the preservation of an ATP-rich state over the impediment of oxidative stress. We are currently investigating this hypothesis in CMV-STOP$^{fl/fl}$-UCP2; GFAP-creER^{T2} mice, where macroglial UCP2 overexpression should impair glaucomatous RGC survival.

Disclosures: D. Hass: None. **C.J. Barnstable:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.12/Z13

Topic: B.12. Glial Mechanisms

Support: RR&D Merit Review #B1097-I

RR&D Career Development Award #IK2-RX001479

NINDS T32-NS043126

Drexel University Interdisciplinary Collaboration and Research Enterprise

Penn's University Research Foundation

NSF #DGE-0947936

Title: Neuronal plasmalemmal damage elicits rapid immune cell activation following diffuse traumatic brain injury in swine

Authors: *K. WOFFORD[1,2,4], K. D. BROWNE[2,4], D. P. BROWN[2,4], M. R. GROVOLA[2,4], J. E. DUDA[3,4], K. L. SPILLER[1], D. CULLEN[2,5]
[1]Drexel Univ., Philadelphia, PA; [2]Ctr. for Brain Injury and Repair, Dept of Neurosurg., [3]Neurol., Univ. of Pennsylvania, Philadelphia, PA; [4]Corporal Michael J. Crescenz Veterans Affairs Med. Ctr., Philadelphia, PA; [5]Ctr. for Brain Injury and Repair, Dept of Neurosurg., Philadelphia, PA

Abstract: It is recognized that disruption of neuronal circuitry from traumatic brain injury (TBI) contributes to acute and persistent symptomology. However, it is not well understood how resident microglia and infiltrating macrophages act to modulate inflammation in the CNS and thereby affect acute neuronal health and homeostasis after TBI. To better elucidate the influence of immune cells on injury progression, we investigated acute immune cell reactivity around injured neurons immediately following TBI. Using a previously characterized closed-head

rotational acceleration injury model in swine, we induced mild or severe TBI and labeled mechanically permeabilized neurons using Lucifer yellow (LY). Acutely after injury (15 minutes), animals were sacrificed and immune cells ($Iba1^+$) were analyzed to investigate their distribution and morphology as a function of distance from neurons exhibiting trauma-induced plasmalemmal disruptions (LY^+). We recently found that regions without permeabilized neurons exhibited no differences in immune cell reactivity in either injured or sham animals. However, activated immune cells were frequently observed adjacent to permeabilized neurons post-injury. Following both mild and severe injuries, immune cell density increased and morphology became more amoeboid in the immediate vicinity of LY^+ neurons in a distance-dependent manner. Interestingly, the increased immune cell density around damaged neurons was not coupled with depletion of immune cell density in LY^- regions, suggesting that additional immune cells may be infiltrating from peripherally-derived monocyte populations. To address this issue, we sought to determine whether acute blood brain barrier compromise and influx of peripheral macrophages contributed to this rapid inflammatory response. Therefore, we examined tissue for evident of IgG, a protein indicative of blood brain barrier disruption. Marginal IgG staining was observed in perivascular regions after TBI. Furthermore, immunohistochemical characterization of CD45, a marker elevated in peripheral monocyte-derived macrophages, was also detected in regions of the brain containing LY^+ neurons suggesting that monocyte-derived macrophages may contribute to this rapid neuroinflammatory response after TBI. Further investigation into the influence of microglia and macrophages on injured neurons could lead to a more robust understanding of neuroinflammation as well as inform the development of immunomodulatory therapeutics.

Disclosures: K. Wofford: None. **K.D. Browne:** None. **D.P. Brown:** None. **M.R. Grovola:** None. **J.E. Duda:** None. **K.L. Spiller:** None. **D. Cullen:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.13/Z14

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Title: Comparison of food intake in newly diagnosed multiple sclerosis patients and healthy individuals- a case-control study

Authors: Y. GHAZAVI[1], Z. BAHADORAN[2], M. NIKFARJAM[2], N. BELADI MOGHADDAM[3], P. MIRMIRAN[2], *M. RAZA[4]
[1]Fac. of Med., [2]Res. Inst. for Endocrine Sci., [3]Dept. of Neurology, Fac. of Med., Shahid Beheshti Univ. of Med. Sci., Tehran, Iran, Islamic Republic of; [4]Baqiyatallah Univ. of Med. Sci., Tehran, Iran, Islamic Republic of

Abstract: Introduction: Nutritional factors considerably affect the incidence, severity of symptoms and progression of MS. However, the exact role of specific nutritional factors and dietary impact of various food items remains unknown in MS. In this study we investigated the differences in the nutritional intake of multiple sclerosis patients and compared the data with the sex and age-matched healthy subjects at selected hospitals in Tehran, IR Iran. Methods: Validated food frequency questionnaire (FFQ) for last one-year dietary assessment was used, in MS patients (n=96) and age matched control group (n=97). Data from FFQ was converted into exact values of micronutrients, calories, food items and food group intake. Finally, the data were compared between MS patients and healthy control subjects by SPSS ver. 18. Odds ratio and 95% confidence interval of multiple sclerosis was calculated in different food groups using multiple logistic regression models adjusted for various potentially confounding variables. Results: There was no significant difference between age (34.62 ±9.68vs. 33.96±8.75) and BMI (23.96 ±4.07 vs. 24.47 ±4.07) of MS and control group respectively. FFQ data showed that MS patients had a lower caloric intake (2631 vs. 2683kcal/d) during previous year, however, the difference was not significant. MS patients had a significant lower intake of sodium (7820±1274 vs. 8395±1592 gm/year), and lycopene (6310±2651 vs. 7615±2712 µm/year), during last year compared with controls. Higher intake of non-processed meat, processed meat, starchy vegetables and sweet snacks increased the odds of multiple sclerosis. ($p<0.03$) There was a 4.3 folds higher Odds for multiple sclerosis in second tertile of processed food consumption than the first tertile. ($p=0.04$) Conclusion: We found higher intake of sodium and lycopene in control group compared to Multiple sclerosis patients. Also, we found that higher intake of non-processed meat, processed meat, starchy vegetables and sweet snacks increased the odds of multiple sclerosis. There seems to be a need for advanced studies to evaluate the probable role of these nutritional factors in pathogenesis of multiple sclerosis and inflammation.

Disclosures: Y. Ghazavi: None. **Z. Bahadoran:** None. **M. Nikfarjam:** None. **N. Beladi Moghaddam:** None. **P. Mirmiran:** None. **M. Raza:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.14/Z15

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Title: Early lead (Pb^{2+}) exposure induces anatomic, ultrastructural and behavioral changes in zebrafish

Authors: L. HUNT[1], F. NIEVES[1], S. NARVAEZ[1], A. MONTEMARANO[1], *S. GUARIGLIA[2,1]

[1]CUNY Col. of Staten Island/Saint Joseph by the Sea High Sch., Staten Island, NY; [2]Col. of Staten Island, Staten Island, NY

Abstract: Exposure to Pb^{2+} during early development has particularly deleterious effects on neurodevelopment that are often permanent. The limbic system is particularly susceptible to toxic insult. Damage to the limbic system can result in the development of aberrant behaviors. Therefore, we sought to determine if early Pb^{2+} exposure led to the development of aberrant behaviors in a zebrafish model and if there were associated histological and ultrastructural changes in the limbic system and associated structures. To address the histological and ultrastructural changes in the limbic system and related structures, we examined brain volume, white matter tracts, and synaptic ultrastructure in the telencephalon of zebrafish exposed to Pb^{2+} during embryogenesis. We found that Pb^{2+} increased the size of the rostral commissure. We did not find any gross anatomical differences in overall size of the brain or any particular brain region. Pb^{2+} did induce an increase in the number of synapses found in the telencephalon. The presynaptic terminal of these synapses exhibited an alteration in vesicular distribution in which vesicular pools were more distant from the presynaptic active zone. Behaviorally, we found that early Pb^{2+} exposure had permanent effects on behavior. Pb^{2+} exposed zebrafish displayed an anxious phenotype in the light-dark box and tank diving tests, impaired learning ability in T-maze and some impairments in social behavior. The sum of our work characterizes novel changes in anatomical and behavioral neurodevelopment that are the result of early Pb^{2+} exposure in a newer and potentially useful model of Pb^{2+} neurotoxicity.

Disclosures: L. Hunt: None. **F. Nieves:** None. **S. Narvaez:** None. **A. Montemarano:** None. **S. Guariglia:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.15/Z16

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH R01 NS092938

University of South Carolina Research Foundation ASPIRE-I

Title: Novel protein targets of dimethyl fumarate in neural cells

Authors: *G. G. PIROLI[1], A. M. MANUEL[1], T. PATEL[1], M. D. WALLA[2], J. WANG[1], L. SHI[4], S. A. LANCI[1], A. GALLOWAY[1], P. I. ORTINSKI[1], D. S. SMITH[3], N. FRIZZELL[1]
[1]Dept Pharmacol/Physiol & Neurosci, Univ. South Carolina Sch. Med., Columbia, SC; [2]Chem.

& Biochemistry, MS Ctr., [3]Biol., Univ. of South Carolina, Columbia, SC; [4]Dept. of Pharmacol. and Toxicology, Med. Col. of Georgia at Augusta Univ., Augusta, GA

Abstract: Fumarate esters including dimethyl fumarate (DMF) have been recently introduced for the treatment of relapsing multiple sclerosis (MS), a chronic inflammatory condition resulting in neuronal demyelination and axonal loss. The proposed mechanism of DMF action involves the depletion of intracellular reduced glutathione and modification of thiols on Keap1, which in turn induces the expression of genes under the control of the antioxidant response element (ARE), including those that facilitate the synthesis and replenishment of cellular glutathione. However, other intracellular thiol residues in neural proteins may also be DMF targets, leading to the irreversible chemical modification of those protein residues by succination. To study the significance of protein succination by DMF in neural cell types, we acutely treated primary rodent neurons and astrocytes, as well as differentiated N1E-115 cells, with 10 to 100 μM DMF, and detected increased succination of a variety of proteins by immunoblotting with an antibody against succinated cysteine residues (2SC) and by mass spectrometry. Using this approach, we confirmed the identification and site of succination of ~25 novel target proteins, including tubulin and cofilin-1. An in vitro functional assay that measures the ability of cofilin-1 to sever the actin cytoskeleton showed that DMF-succinated cofilin-1 loses some activity, potentially inhibiting cofilin's cytoskeletal remodeling activity, an effect that could be beneficial in the modulation of myelination during MS. In addition, the oxygen consumption rate of N1E-115 cells measured with a Seahorse XF24 analyzer, as well as the expression of mitochondrial proteins related to the mitochondrial energy production, were only slightly affected by the highest doses of DMF, indicating a low probability of DMF toxicity on neural cells. In summary, we demonstrate that in addition to the stimulation of the antioxidant response, DMF increases protein succination in neural cells with a potential blockade of myelin loss and limited adverse effects.

Disclosures: G.G. Piroli: None. **A.M. Manuel:** None. **T. Patel:** None. **M.D. Walla:** None. **J. Wang:** None. **L. Shi:** None. **S.A. Lanci:** None. **A. Galloway:** None. **P.I. Ortinski:** None. **D.S. Smith:** None. **N. Frizzell:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.16/Z17

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: Research Sector OMICSRU grant SRUL02/13.

Title: Neuroprotective effect of thymoquinone on cuprizone-induced multiple sclerosis in mice

Authors: *W. M. RENNO[1], H. L. SADEK[2]
[2]Anat., [1]Kuwait Univ., Safat, Kuwait

Abstract: Multiple sclerosis (MS) is a chronic inflammatory disease in which the myelin sheaths surrounding the axons of the CNS are damaged, leading to demyelination and scarring as well as a broad spectrum of symptoms that are considered one of the leading causes of disability in the developed world. The exact cause of MS is still unknown, but some data indicate that it likely occurs as a result of some combination of genetic, environmental and infectious factors. Several studies have shown that *Nigella sativa* and its active ingredient, thymoquinone, have neuroprotective effects against many degenerative diseases. A strong association exists between thymoquinone and the fundamental biological processes that facilitate regeneration, such as anti-oxidant, anti-inflammatory, and anti-apoptotic effects. The aim of this study is to investigate the role of thymoquinone in the neuroprotection and remyelination following cuprizone (CPZ)-induced MS mouse model. **Methods:** Thirty C57BL/6J mice were randomly divided into three groups: (I) Control given normal mouse chow, (II) CPZ+saline-treated and (III) CPZ+thymoquinone-treated groups. The experimental groups will be given 0.2% CPZ in a milled mouse chow mix *ad libitum* for five weeks. The function and severity of the CNS impairment were assessed and the motor neurobehavioral skills were evaluated. The salvaged brains were assessed for histopathological and immunohistochemistry Changes. **Results:** Thymoquinone-treated group showed significant ($P<0.0001$) motor improvement in the rotarod test following two weeks of treatment compared with CPZ-saline-treated mice. A remarkable increase of the Luxol fast blue staining intensity in the corpus callosum (CC) area was observed with a marked increase in cellularity in the thymoquinone-treated animals compared to saline-treated controls. Ultrastructural analysis of the CPZ+Saline mice showed heterogeneous myelin sheaths surrounding axons in the CC with noticeable loose myelin sheaths and few lamella. Further, oligodendrocytes showed apoptotic changes such as condensed and fragmented nuclei compared. In contrast, the thymoquinone-treated animals showed a decrease in apoptotic cell appearance with a remarkable increase in the myelin sheaths and lamellae and oligodendrocytes. **Summary:** Thymoquinone-treated mice showed a faster regenerative morphology and remyelination after two weeks of stopping CPZ administration in contrast to CPZ saline-treated mice. Thymoquinone neuroprotective properties may be useful adjunct therapeutic regimen to alleviate neurobehavioral motor symptoms as well as speed up the remyelination process in the MS mice model.

Disclosures: W.M. Renno: None. **H.L. Sadek:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.17/Z18

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: Ministero della Salute GR-2011-02347036

Ministero della Salute GR-2011-02351422

Biogen Idec

Title: Neuroprotective effect of monomethyl fumarate on inflammation-driven synaptopathy in a MS preclinical model

Authors: *D. FRESEGNA[1,2], F. DE VITO[1,2], A. MUSELLA[3], A. GENTILE[1,3], S. BULLITTA[1,2], F. RIZZO[1,2], A. PAOLILLO[4], D. CENTONZE[1,2], G. MANDOLESI[3]
[1]Medicina dei sistemi, Univ. of Rome Tor Vergata, Roma, Italy; [2]IRCCS Inst. Neurologico Mediterraneo (INM) Neuromed, Pozzilli (IS), Italy; [3]Lab. of Neuroimmunology and Synaptic Palsticity, IRCCS Fondazione Santa Lucia, Roma, Italy; [4]Biogen, Rome, Italy

Abstract: Excitotoxic synaptopathy is emerging as an early pathophysiological hallmark of multiple sclerosis (MS) and of its mouse model, experimental autoimmune encephalomyelitis (EAE). It includes both structural and functional synaptic dysfunctions induced by inflammation, typically resulting in a marked imbalance of glutamatergic and GABAergic transmissions. In the long-term, excessive synaptic excitation causes neuronal damage, leading to motor and cognitive dysfunctions.
Recently, we highlighted new crucial molecular players linking inflammation and altered glutamate transmission in EAE cerebellum. We demonstrated that both the proinflammatory cytokine interleukin 1 beta (IL-1b) and the microRNA miR-142-3p are expressed in resident as well as infiltrating cells, and participate in a regulatory axis responsible for the downregulation of the glutamate aspartate transporter/excitatory amino acid transporter 1 (GLAST/EAAT1). Inflammation-driven GLAST/EAAT1 reduction compromises the glutamate reuptake from the synaptic cleft and causes increased duration of excitatory currents recorded from cerebellar neurons.
Since EAE/MS synaptopathy is precocious and potentially reversible, it represents an attractive therapeutic target. Thus, we asked whether EAE synaptopathy could be directly targeted by disease-modifying treatments, like the oral drug dimethyl fumarate (DMF) which metabolizes to active metabolite monomethyl fumarate (MMF).
Our e*x vivo* experiments in EAE cerebellar slices showed that acute incubation MMF was able to correct the kinetic abnormalities of glutamatergic currents recorded from EAE Purkinje cells. Mechanistically, we provide evidence that MMF can reestablish glutamate homeostasis by restoring the level of the glial glutamate transporter GLAST/EAAT1, through the reduction of the expression of miR-142-3p, without affecting IL-1b mRNA level.
Consistently with ex vivo incubation of MMF in EAE cerebellar slices, therapeutic intracerebroventricular administration of MMF in EAE mice, given in the symptomatic phase of the disease, was able to mitigate cerebellar synaptic dysfunctions.
Overall, our findings highlighted a novel direct neuroprotective mechanism through which MMF is able to restore glutamate homeostasis in EAE cerebellum and, thus, to prevent glutamate-

dependent excitotoxic damages, by perturbing the detrimental IL-1b-miR-142-3p-GLAST/EAAT1 regulatory axis typical of the EAE cerebellum.

Disclosures: D. Fresegna: None. **F. De Vito:** None. **A. Musella:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Ministero della Salute. **A. Gentile:** None. **S. Bullitta:** None. **F. Rizzo:** None. **A. Paolillo:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Biogen. **D. Centonze:** None. **G. Mandolesi:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Ministero della Salute.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.18/Z19

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: MS Society of Canada

Canadian Institutes of Health Research

Title: Copper-iron connection in cuprizone-induced oligodendrocyte loss

Authors: *P. JHELUM, E. SANTOS-NOGUEIRA, S. DAVID
The Res. Inst. of the McGill Univ., Montreal, QC, Canada

Abstract: The copper chelator, cuprizone (CZ), induces oligodendrocyte (OL) death and demyelination in the CNS, which is seen particularly well in the corpus callosum. Copper chelation would reduce availability of copper-containing enzymes involved in cellular iron efflux. Therefore, CZ treatment can be expected to alter iron homeostasis in OL and lead to iron accumulation and cell death. As iron is needed for remyelination, increased iron in macrophages may be beneficial during the remyelination phase. In this study, we assessed the loss of mature OL and oligodendrocyte precursor cells (OPCs) from 2 to 5 weeks after start of CZ treatment, and correlated this with changes in expression of ferritin, a surrogate marker for cellular iron. Mature CC1+ OL showed a sharp loss at 2 and 4 weeks suggesting two cycles of cell death. The loss of OL at 2 weeks is correlated with a reduction in the iron efflux protein, hephaestin, which is expressed in OL. The number of OPCs showed loss at 2 weeks followed by a gradual increase

of ~ 2-fold at 5 weeks. Densitometry analysis showed ferritin labeling (sign of iron overload) increased ~2 - 3-fold at 2 and 3 weeks, a sharp reduction at 4 weeks followed by a second phase increase by 3-fold at 5 weeks. The first increase in ferritin occurs in OL, and the second increase occurs in macrophages. These results suggest that loss of copper can lead to iron-mediated OL death at the early time points, and the later increase in macrophages may have a role in remyelination.

Disclosures: P. Jhelum: None. **E. Santos-Nogueira:** None. **S. David:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.19/Z20

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: NMSS Grant RG5022A1

Title: The retinoic acid synthesizing enzyme, retinaldehyde dehydrogenase 2 (RALDH2), regulates mouse CNS remyelination

Authors: *S. E. NANESCU, N. WATHIEU, L. ROSKO, D. CHA, J. K. HUANG
Biol., Georgetown Univ., Washington, DC

Abstract: Multiple sclerosis (MS) is an autoimmune-driven neurodegenerative disease characterized by chronic inflammation, demyelination and axonal injury. Retinoic acid (RA), the active form of Vitamin A, is a key signaling molecule in tissue regeneration in many species. Before becoming RA, retinol is first oxidized to retinaldehyde by retinol dehydrogenases, and oxidized further by retinaldehyde dehydrogenases (RALDHs). Among these RALDHs, the most abundantly expressed during development is RALDH2. The objectives of the study were to investigate RALDH2 expression during remyelination and to determine whether local RA synthesis in demyelinating lesions occurs to promote CNS remyelination. In order to assess the role of RA in CNS remyelination, an acute, focal demyelinated lesion was generated by injecting 1% lysolecithin into the mouse spinal cord, and we have used a retinoic acid response element (RARE)-reporter mouse, a mouse that expresses beta-galactosidase (*lacZ*) gene under the control of RARE. As a result, we have found active RA signaling within the remyelinating lesion. To investigate specifically the role of RALDH2 in remyelination, focal demyelination was performed on a tamoxifen inducible RALDH2 knockout mouse line. We found that following demyelination, the Raldh2 conditional KO mice displayed significantly increased pro-inflammatory macrophages in the lesion. Moreover, we found a significant reduction in in the number of oligodendrocyte progenitor cells (OPCs) and mature oligodendrocytes in the lesion. The reduction in OPCs and oligodendrocytes ultimately resulted in decreased remyelination, as

shown by MBP immunohistochemistry quantification. Furthermore, we found that the injection of RA into lesions is able to rescue progenitor and oligodendrocyte numbers in RALDH2 conditional knockout mice by 14 days after injury. Together, these results suggest that RALDH2 is necessary for RA synthesis following CNS demyelination, and that RA production in lesions modulates inflammation and increases the pool of OPCs to promote remyelination.

Disclosures: S.E. Nanescu: None. **N. Wathieu:** None. **L. Rosko:** None. **D. Cha:** None. **J.K. Huang:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.20/Z21

Topic: H.01. Animal Cognition and Behavior

Support: NIDCR T32 DE014320

American Surgical Association Foundation GRT00035777

Title: Resident microglia form rod-shaped structures in cortical regions of neuronal damage and inflammation after diffuse traumatic brain injury

Authors: *K. G. WITCHER[1], B. N. BENNER[2], D. B. MCKIM[2], J. LIFSHITZ[4], D. S. EIFERMAN[3], J. P. GODBOUT[5]
[1]Dept. of Neurosci., Ohio State Univ., Columbus, OH; [3]Surgery, [2]The Ohio State Univ., Columbus, OH; [4]Barrow Neurolog. Inst., Phoenix Children's Hosp., Phoenix, AZ; [5]Neurosci., Ohio State Univ. Dept. of Neurosci., Columbus, OH

Abstract: Traumatic brain injury (TBI) elicits immediate neuroinflammation that contributes to acute cognitive, motor, and affective disturbances. Although acute impairments resolve after mild to moderate TBI, inflammatory processes persist and may underlie neuropsychiatric and cognitive complications that arise long after injury. Microglia, the innate immune cells resident to the brain, are key mediators of acute and chronic inflammation. After diffuse brain injury, microglia form elongated, rod-like structures in the cerebral cortex. While rod-shaped cells have been previously shown to peak in number 7 days post-injury, their derivation and role in pathophysiology is unclear. We first aimed to determine the origin of rod-shaped cells. We found that both rod microglia and nearby activated microglia upregulated CD45. Using GFP bone marrow chimeras, we confirmed that these CD45+ cells were indeed resident CNS microglia. BrdU labeling confirmed that the rod cells were resident microglia and not the result of TBI-induced microglial proliferation. We also found that some rod-shaped microglia aligned with blood vessels, but the frequency was comparable to naïve mice, suggesting that microglia align

with the vasculature at baseline. Here we also show novel data using Thy1-YFP-H transgenic mice that rod microglia were in close proximity to axotomized (ATF-3+) neurons and aligned with apical pyramidal dendrites. Regions with more rod microglia also had increased astrocyte reactivity, as demonstrated by increased GFAP labeling. Genes associated with neuronal injury (CSF-1, ATF-3) and immune reactivity (CCL-2, MHCII, TREM2) are upregulated in cortical homogenate both 3 and 7 dpi. Together, these findings suggest that rod microglia, which were present at the time of injury, form rod-shaped structures in regions susceptible to neuronal injury and are a part of a larger inflammatory response following diffuse TBI. Future studies aim to further delineate the gene expression of rod microglia, nearby activated microglia, injured neurons, and reactive astrocytes following brain injury.

Disclosures: K.G. Witcher: None. **B.N. Benner:** None. **D.B. McKim:** None. **J. Lifshitz:** None. **D.S. Eiferman:** None. **J.P. Godbout:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.21/Z22

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: Danish Strategic Research Council (COGNITO)

HEALTH-F2-2011-278850 (INMiND)

Desirée and Niels Ydes Foundation

Title: Add insult to injury: microglia activation in neuropsychiatric diseases?

Authors: *C. K. DONAT[1,2], M. SHUKUROGLOU[1], D. R. OWEN[1], L. H. PINBORG[2,3], M. SASTRE[1], J. D. MIKKELSEN[2]
[1]Div. of Brain Sci., Imperial Col. London, London, United Kingdom; [2]Neurobio. Res. Unit, [3]Epilepsy Clin., Copenhagen Univ. Hosp. - Rigshospitalet, Copenhagen, Denmark

Abstract: Several studies implicated the immune response in neuropsychiatric diseases, e.g. schizophrenia and depression. In the brain, immune crosstalk can result in a microglial response, detectable with radioligands for the 18 kDa Translocator Protein (TSPO). However, *in vivo* molecular imaging studies with TSPO tracers have yielded inconsistent results, reporting no changes, decreases or increases when comparing patient populations to controls. To further study the role of TSPO in these diseases, we chose a well-defined cohort (Stanley Medical Research Institute, Neuropathology consortium) including subjects with schizophrenia (SCZ), Bipolar (BP) and Major depressive disorder (MDD), and controls (HC, n=15 each), genotyped for rs6971 single-nucleotide polymorphism. Binding of three TSPO tracers, [^{3}H]PK11195, [^{123}I]CLINDE

and [^{3}H]PBR28 was assessed by autoradiography in consecutive sections from the orbitofrontal cortex and quantified in cortical grey (GM) and white matter (WM).
In MDD, mean specific [^{3}H]PBR28 binding was higher in patients than HC in GM (+45%, p=0.03), and a trend was also observed in WM (+35%, p=0.08). In BP and SCZ, although mean specific [^{3}H]PBR28 binding was higher in both GM (BP: 18%, SCZ: 23%) and WM (BP: 9%, SCZ: 22%), these differences were not significant. Specific binding of [^{3}H]PK11195 and [^{123}I]CLINDE was similar to HC in all 3 diseases, [GM (-2% to +12%) and WM (-8% to +3%)] with no significant differences. Nonspecific binding was different between the tracers, very low in [^{3}H]PBR28 (12%) and higher for [^{3}H]PK11195 (44%) and [^{123}I]CLINDE (57%). Across all subjects, [^{123}I]CLINDE and [^{3}H]PBR28 binding was positively correlated in GM and WM (Pearson's r=0.384, 0.429; p<0.01). [^{3}H]PK11195 binding was weakly correlated with [^{123}I]CLINDE in GM (r=0.269, p=0.03) but not WM and not with [^{3}H]PBR28 (GM: p=0.056, WM: p=0.051).
The findings show a general increase of TSPO in neuropsychiatric diseases when using [^{3}H]PBR28. No differences are found using [^{123}I]CLINDE or [^{3}H]PK11195 and binding properties of the radioligands may account for this discrepancy. The increase in [^{3}H]PBR28 binding was significant in GM of MDD patients, indicating a microglia response and supporting further *in vivo* TSPO PET studies in patients with depression.

Specific binding of three structurally different TSPO radioligands in the SMRI Neuropathology cohort

	rs6971 SNP	**[^{3}H]PK11195**				**[^{123}I]CLINDE**				**[^{3}H]PBR28**			
	# of HAB:MAB:LAB	**fmol/mg TE**	**SD**	**% of HC**	**p-value vs HC**	**fmol/mg TE**	**SD**	**% of HC**	**p-value vs HC**	**fmol/mg TE**	**SD**	**% of HC**	**p-value vs HC**
Grey matter													
Healthy controls (HC)	9:6:0	76.3	15.9			55.6	17.7			58.9	16.2		
Bipolar disorder (BP)	7:7:1	85.2	17.4	112	0.16	58.4	11.2	105	0.62	69.3	35.0	118	0.53

Major Depressive disorder (MDD)	9:6:0	75.1	26.7	98.4	0.88	58.3	13.0	105	0.64	85.1	42.3	145	*0.03*
Schizophrenia (SCZ)	9:5:1	78.3	18.7	103	0.47	56.0	8.97	101	0.94	72.5	34.6	123	0.18
White matter													
Healthy controls (HC)	9:6:0	54.2	15.5			56.6	19.0			28.5	9.24		
Bipolar disorder (BP)	7:7:1	51.3	18.3	94.7	0.65	54.3	8.68	96.0	0.68	31.0	16.5	109	0.99
Major Depressive disorder (MDD)	9:6:0	55.7	28.6	103	0.85	58.2	14.9	103	0.80	38.5	19.2	135	0.08
Schizophrenia (SCZ)	9:5:1	49.6	19.0	91.5	0.47	57.5	10.4	102	0.87	34.8	17.3	122	0.23
Specific binding expressed as mean fmol/mg tissue equivalents, significance tested with unpaired t-test or Mann-Whitney test, n=15 for each group. HAB: high-affinity binder, MAB: mixed-affinitybinder, LAB: low-affinity binder													

Disclosures: **C.K. Donat:** None. **M. Shukuroglou:** None. **D.R. Owen:** None. **L.H. Pinborg:** None. **M. Sastre:** None. **J.D. Mikkelsen:** None.

Poster

390. Glia-Neuronal Communication in Health and Disease

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 390.22/Z23

Topic: B.08. Synaptic Plasticity

Support: 5T32D046388-09

Title: Neuroglia glutamate signaling in the adult brain is dependent on mGluR3

Authors: *N. A. SMITH[1], ***N. A. SMITH**[1], L. K. BEKAR[2], J. LIU[1], M. NEDERGAARD[3], V. GALLO[1]
[1]Ctr. for Neurosci. Res., Children's Natl. Hlth. Syst., Washington, DC; [2]Dept. of Pharmacol., Univ. of Saskatchewan Saskatoon SK, Saskatoon, SK, Canada; [3]Ctr. for Translational Neuromedicine, Univ. of Rochester Sch. of Med. and Dent., Rochester, NY

Abstract: The concept of neuroglia signaling in the adult brain has come under question because recent studies have shown that astrocytic mGluR5 is developmentally regulated and is undetectable after postnatal week 3. Thus, ruling out the possibility for glutamate evoked Ca^{2+} signaling in mature astrocytes. However, astrocytes express another major metabotropic glutamate receptor which is mGluR3. Unlike mGluR5, mGluR3 is a Gi couple G-protein receptor that is expressed throughout adulthood. Although mGluR3 activation is known to inhibit adenylate cyclase, new studies have revealed that mGluR3 can also induce intracellular Ca^{2+} changes but the mechanism behind this induction has not been fully explored in astrocytes. To explore mGluR3 Ca^{2+} induction in astrocytes, we employed a new transgenic mouse line that expresses a *Polr2a* based Cre-dependent GCaMP5G Ca^{2+} indicator that has also been tagged with IRES-tdTomato (tdTm) to aid detection of positive cells. These animals have been crossed with an inducible Cre line (GFAPcreER) that drives specific expression of GCaMP5G-tdTm in numerous astrocytes and enables monitoring of intracellular Ca^{2+} dynamics in astrocytes throughout the CNS using 2-photon microscopy. After determining mGluR3 significance in astrocytic Ca^{2+} dynamics, next we evaluated whether mGluR3 evoked Ca^{2+} transients regulate astrocytic modulation of a faster form of synaptic activity know as Transient Heterosynaptic Depression (tHSD). This study redefines the mechanism of neuroglia signaling in the adult brain and places astrocytes as key players in informational processing.

Disclosures: **N.A. Smith:** None. **L.K. Bekar:** None. **J. Liu:** None. **M. Nedergaard:** None. **V. Gallo:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.01/Z24

Topic: C.07. Ischemia

Support: NIH

NSFC

Title: Synergistic role of endogenous and exogenous 17β-estradiol in protection of ischemic neurons

Authors: *R. WANG
MEDICAL RECEARCH CENTER, Norteh CHINA UNIVERSITY OF SCIENCE AND TEHCNOLOGY, TANGSHAN, China

Abstract: Recent studies from our laboratory revealed that brain-derived E2 (BDE2) is elevated in the hippocampal CA1 region after global cerebral ischemia (GCI), and that it exerts a neuroprotective effect, although the mechanisms underlying the neuroprotective effect were unclear. The goals of the current study were to 1) examine the mechanisms underlying BDE2 neuroprotection after GCI, 2) examine whether exogenous E2 regulates aromatase/BDE2, and determine whether BDE2 is required for exogenous E2-mediated neuroprotection. To address these goals, aromatase/BDE2 was inhibited by either central administration of an aromatase inhibitor, letrozole or aromatase antisense oligonucleotide (AS) in ovariectomized adult female rats with or without exogenous E2 following GCI. The results showed that aromatase expression initially decrease at 3h, rebound to basal level at 1d, and then is robustly elevated at 3 d and 7d reperfusion in the hippocampal CA1 region after CGI. Administration of exogenous E2 increased aromatase expression in the hippocampal CA1 region at reperfusion 3h, 1d and 3d. Interestingly, immunofluorescent staining showed that aromatase was primarily expressed in neurons in sham and E2 treatment groups; however, at reperfusion 3d and 7d aromatase was predominantly expressed in reactive astrocytes. Intriguingly, central administration of aromatase AS or letrozole abolished both the GCI and exogenous E2-induced enhancement of aromatase at 7d reperfusion. Furthermore, letrozole and aromatase-AS treatment attenuated both endo- and exogenous E2-induced phosphorylation/activation of the prosurvival factors, ERK1/2 and CREB, as well as synaptic protein expression of PSD95 and synaptophysin after GCI. Taken as a whole, the results suggest that the neuroprotective effects of BDE2 after GCI involve enhancement of prosurvival ERK and CREB signaling and enhanced synaptic plasticity in the hippocampus. Furthermore, BDE2 appears to exert a “priming effect” that is critical for the beneficial neuroprotective effects of exogenous E2 after GCI. (This research was supported by NIH/NINDS research grant R01NS088058 to Dr. DW Brann and National Natural Science Foundation of China, 81671223 to Dr. RM Wang)

Disclosures: R. Wang: None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.02/Z25

Topic: C.07. Ischemia

Support: NIH/NIGMS Grant T32 GM007507

NIH Grant R01-NS37570

AHA 1525500022

NIH Grant R01-HL128778

Title: Myeloid cell produced VEGF-A contributes to acute ischemic stroke damage

Authors: *A. RAYASAM[1], A. LINDSTEDT[1], J. KIJAK[1], T. KIM[1], M. HSU[1], M. HERBATH[1], J. HARDING[2], A. NAGY[2], R. VEMUGANTI[1], M. SANDOR[1], Z. FABRY[1]
[1]Univ. of Wisconsin - Madison, Madison, WI; [2]Lunenfeld-Tanenbaum Res. Institute, Mount Sinai Hosp., Toronto, ON, Canada

Abstract: Stroke is a widespread disease that affects approximately 15 million people globally every year. During ischemic stroke, secondary damage occurs due to immune cell-mediated reperfusion. Myeloid cells, phagocytic immune cells, are responsible for debris clearance and inflammation during the early stages of reperfusion after acute ischemic stroke, but the molecular mechanisms for how these cells contribute to stroke damage is still unclear. While vascular endothelial growth factor-A (VEGF-A) is known to influence the blood-brain barrier (BBB), limited information is known about how myeloid cell produced VEGF-A contributes to the immune responses after stroke. Using the transient middle cerebral artery occlusion (tMCAO) in mice, we show that myeloid cells are recruited to the ischemic brain rapidly during reperfusion and that a high proportion of these cells produce VEGF-A. Furthermore, we utilize transgenic murine models, which alter levels of VEGF-A and target myeloid cell produced VEGF-A specifically, to test how VEGF-A contributes to the acute phase of ischemic stroke. Overall, these data provide novel information for how VEGF-A contributes to stroke pathology.

Disclosures: **A. Rayasam:** None. **A. Lindstedt:** None. **J. Kijak:** None. **T. Kim:** None. **M. Hsu:** None. **M. Herbath:** None. **J. Harding:** None. **A. Nagy:** None. **R. Vemuganti:** None. **M. Sandor:** None. **Z. Fabry:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.03/Z26

Topic: C.07. Ischemia

Title: ASIC1a mediates autophagy activation and neuronal death induced by ischemic brain injury mice model

Authors: *X. SUN
The First Affiliated Hosp. of Soochow Universit, Jiangsu, China

Abstract: Tissue acidification and cell autophagy in the early days of the ischemic brain injury plays an important role, organization acidification can lead to acid sensitive ion channels (ASICs) abnormal activation. Our previous work showed that ASICs activation could induce the change of autophagy activity and lead to the death of neurons. However, it is unclear that the mechanism of ASIC1a and autophagy participated in the pathological process of ischemic brain injury. In this study, we constructed the middle cerebral artery ligation mice brain ischemia model at different time points and checked the expression of ASIC1a and autophagy activity by Western blot. The results showed that ASIC1a was activated in the ischemic brain damage and expression of autophagy related protein LC3-II was highest increased with p62 was decrease after ischemia 24 h, suggesting that ASIC1a and autophagy were activated in the process of neuronal damage after cerebral ischemia in mice. Furthermore, to investigate whether the autophagy activity was induced by ASIC1a, we constructed the ischemia model in wild-type mice, ASIC1a overexpression, and ASIC1a knockout mice then detected the autophagy activity and neuronal death by Nissl staining. We found that autophagy and neuronal death were induced in ASIC1a overexpression group higher than wild-type and ASIC1a knockout mice, so ASIC1a mediates autophagy increased the activity of neuronal death in the brain after ischemic treatment. Moreover, to verify the role of ASIC1a in the autophagic death pathway of neuronal damage after ischemia in vitro, the primary cultured neurons were treated with hypoxia and acid. The results showed that the expression of ASIC1a and autophagy pathway were increased, and ASIC1a regulated autophagy and neuronal death in vitro. In summary, this is the first report of autophagy pathway and neuronal death were regulated by ASIC1a activation induced by ischemic brain damage, which discuss ASIC1a in ischemic brain injury mechanism of the role of autophagy death pathway, providing a new method to improve the ischemic brain damage and theoretical basis.

Disclosures: X. Sun: None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.04/Z27

Topic: C.07. Ischemia

Support: AHA Scientist Development Grant 16SDG31500001

NARSAD Young Investigator Grant 25369

NIH Grant NS46742

Title: Global ischemia activates miR-34b/c mediated by p53 in hippocampal neurons destined to die

Authors: *J.-Y. HWANG, F. PONTARELLI, B. COURT VAZQUEZ, S. ZUKIN
Dept Neurosci, Albert Einstein Col. Med., Bronx, NY

Abstract: Transient global ischemia arising as a consequence of cardiac arrest in humans causes selective, delayed death of hippocampal CA1 pyramidal neurons and cognitive impairment. Effective treatments to ameliorate the neurodegeneration and cognitive dysfunction associated with global ischemia are an unmet need. Emerging evidence points to a widespread role for microRNAs (miRNAs) as key modulators of target gene expression in neurons. Accordingly, dysregulation of miRNAs are implicated in the pathophysiology of neurodegenerative disease and neurological disorders. Our findings, derived *via* miRNA-seq, indicate that a subset of microRNAs is altered in postischemic CA1 including miR-34b/c. Dysregulation of miR-34 has been implicated in pathophysiology of neurological disorder such as Parkinson's disease and epilepsy. However, a role for miR-34 in the pathogenesis of global ischemia is, as yet, unclear. Here we show ischemia induces p53-dependent activation of miR-34b/c and downregulation of its target genes, which together promote neuronal death in selectively vulnerable hippocampal CA1 *in vivo*. Consistent with this, inhibition of miR-34b/c affords neuroprotection in ischemia. These findings document a causal role for p53-dependent activation of miR-34b/c in neuronal death and identify a novel therapeutic target for amelioration of the neurodegeneration and cognitive deficits associated with ischemic stroke.

Disclosures: J. Hwang: None. **F. Pontarelli:** None. **B. Court Vazquez:** None. **S. Zukin:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.05/Z28

Topic: C.07. Ischemia

Support: Department of Health, James & Esther King Biomedical Grant 09KW-11

Department of Health, James & Esther King Biomedical Grant 6JK08

Title: Granulocyte-colony stimulating factor induced neuroprotection in rodent models of focal and global cerebral ischemia

Authors: *J. M. MENZIE-SUDERAM, J. MODIE, H. CHOU, P. TRUJILLO, K. MEDLEY, R. TAO, H. PRENTICE, J.-Y. WU
Biomed. Sci., Florida Atlantic Univ., Boca Raton, FL

Abstract: We have previously shown that DETC-MeSO, Sulindac, and G-CSF could result in brain protection from ischemic injury. In this report we further investigate the mechanisms underlying the protection by G-CSF in terms of preventing ischemic damage caused by ER stress, mitochondrial dysfunction and autophagy. Using the middle cerebral artery occlusion (MCAO) model in rat we have established that G-CSF treatment protected against focal ischemia by reducing endoplasmic reticulum (ER) stress induced apoptosis and preserving the ER integrity. G-CSF treatment significantly attenuated the expression of key proteins involved in the ER stress induced apoptosis pathway; ATF4, ATF6, p-p38MAPK and CHOP as well as reducing the level of the intraluminal ER stress sensor, GRP78. In this model, G-CSF also reduced the general cellular stress marker HSP27. Similar alterations have been found in the BCAO mouse model of global ischemia targeting ER stress mechanisms. Furthermore we have shown altered expression caused by ischemia in the BCAO model in expression of the mitochondrial markers Drp1 and Opa1 as well as the autophagy marker Beclin. Expression of these markers was restored to sham non - ischemic control levels by G-CSF treatment. In conclusion this G-CSF mechanism-based therapy was found to afford ischemic protection through preventing tissue damage at the level of ER-, mitochondrial-, and autophagy related signaling events.

Disclosures: J.M. Menzie-Suderam: None. **J. Modie:** None. **H. Chou:** None. **P. Trujillo:** None. **K. Medley:** None. **R. Tao:** None. **H. Prentice:** None. **J. Wu:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.06/Z29

Topic: C.07. Ischemia

Title: Molecular analysis of mutant high-temperature requirement serine protease a1 identified in patients with familial cerebral small vessel disease

Authors: *M. UEMURA[1], H. NOZAKI[2], T. KATO[3], A. KOYAMA[3], O. ONODERA[4]
[1]Brain Res. Inst. (BRI), Niigata Univ., Niigata, Japan; [2]Niigata Univ., Niigata, Japan; [3]Niigata Univerity, Niigata, Japan; [4]Brain Res. Inst. (BRI), Niigata Univerity, Niigata, Japan

Abstract: Background: Cerebral autosomal recessive arteriopathy with subcortical infarcts and leukoencephalopathy (CARASIL), which is a rare familial cerebral small vessel disease (CSVD), is caused by mutations in high-temperature requirement serine protease A1 (HTRA1). Although it has been believed that mutations in HTRA1 cause CARASIL only in a recessive manner,

recent reports suggested that some specific mutations in HTRA1 cause CSVD even in a heterozygous state (Hetero). HTRA1 mutations in Hetero cause CSVD via dominant negative effects, on the other hand, HTRA1 mutations in homozygous or compound heterozygous states (Homo) cause CSVD via loss of HTRA1 protease activity. It has been remained unknown what type of mutated HTRA1 proteins have pathogenicity in Hetero. The aim of this study to investigate the pathogenicity of previously reported mutations in HTRA1 in Hetero.
Methods: Five mutations reported as Homo (R166C, A173T, G295R, A321T, L364P) and 10 mutations in HTRA1 reported as Hetero (S121R, A123S, R133G, R166L, A173P, S284G, S284R, P285Q, F286V, D450H) were analyzed. First, the protease activity of each purified wild type HTRA1 (WT) and mutant HTRA1 proteins was measured by using fluorescein isothiocyanate-labeled casein as a substrate. Second, the protease activity of purified protein from co-expression of WT and each mutant HTRA1 was evaluated. Purified protein of S328A, which was artificially loss of protease activity, was used as a negative control. In addition, HTRA1 oligomeric structure of each mutant protein was analyzed by size-exclusion chromatography system.
Results: The protease activities of 4 mutations in Hetero were similar or higher as compared with those of WT. The other 6 mutations in Hetero (S121R, R166L, A173P, S284R, P285Q, A321T) and all 5 in Homo revealed significantly lower protease activities than those of WT ($p < 0.05$). Using purified protein from co-expression of WT and each mutant, the protease activities of R166L/WT, A173P/WT, S284R/WT, A173T/WT and G295R/WT were significantly decreased as compared with WT/S328A ($p < 0.05$). The size-exclusion chromatography showed that S284R was eluted as a trimer like S328A and WT. The other 4 mutations (R166L, A173P, A173T and G295R) were eluted as a monomer.
Conclusion: Three of 10 mutations reported in Hetero and 2 of 5 HTRA1 mutations in Homo have dominant negative effects. These HTRA1 mutations might cause CSVD even in a heterozygote state. There is a possibility that the other mutants do not contribute to the pathogenesis of CSVD. The significance of the mutations for the pathogenesis of CSVD should be carefully evaluated by the biochemical analysis.

Disclosures: **M. Uemura:** None. **H. Nozaki:** None. **T. Kato:** None. **A. Koyama:** None. **O. Onodera:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.07/Z30

Topic: C.07. Ischemia

Support: NIH Grant NS073779

Title: Potential role of endoplasmic reticulum stress in recurrent hypoglycemia-induced increase in ischemic brain damage

Authors: *A. K. REHNI[1], V. SHUKLA[2], K. R. DAVE[2]
[2]Neurol., [1]Univ. of Miami Sch. of Med., Miami, FL

Abstract: Diabetes is a serious metabolic disease and stroke among diabetics is noted to be associated with wide spread brain damage. Anti-diabetic drug therapy related episodes of hypoglycemia cause hypoglycemia associated autonomic failure and eventually lead to development of recurrent hypoglycemia (RH). Our laboratory have previously reported that prior exposure of RH exacerbates ischemic brain injury in insulin-treated diabetic (ITD) rats. However, mechanisms known to cause this injury are least understood. Acute hypoglycemia activates unfolded protein response in liver. Glucose starvation also activates endoplasmic reticulum (ER) stress. Cerebral ischemia activates one of three ER stress pathways namely protein kinase RNA-like endoplasmic reticulum kinase (PERK) -eukaryotic initiation factor 2α (eIF2α) pathway. However, the role of ER stress in RH-induced aggravation of ischemic brain damage among treated diabetics is not known. Therefore, aim of the study was to evaluate post-cerebral ischemic ER stress in ITD rats exposed to RH. We determined levels of total and phospho-PERK, and C/EBP homologous protein (CHOP) (using western blot analysis) as markers of ER stress in Naïve (n=6), ITD (n=5), and ITD + RH (representing RH exposed treated diabetic) (n=8) groups. Rats were rendered diabetic by administration of streptozotocin and 2-3 weeks later, insulin pellet implantation was done to treat diabetic hyperglycemia. After 2-3 weeks, sub-cutaneous injection of insulin was given to induce hypoglycemia for 3 hours per day for 5 consecutive days. Overnight after last hypoglycemia exposure, global cerebral ischemia was induced by bilateral carotid artery occlusion with hypotension for eight minutes. We observed that pPERK to total PERK ratio in the ITD + RH group was higher by 57% ($P<0.05$), and 36% ($P<0.05$) as compared with naïve and ITD groups, respectively. We found that the level of CHOP in the ITD + RH group was higher by 30% ($P<0.05$), and 59% ($P<0.05$) as compared with naïve and ITD groups, respectively. Thus, we conclude that cerebral ischemia increases ER stress in RH-exposed ITD rats and may play a role in increased cerebral ischemic damage observed in RH exposed ITD rats. Confirming the role of ER stress in RH-induced aggravation of ischemic brain damage may help in developing new therapeutic options for diabetic stroke patients.

Disclosures: **A.K. Rehni:** None. **V. Shukla:** None. **K.R. Dave:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.08/Z31

Topic: C.07. Ischemia

Support: NIH grant HL038206

NIH grant NS079331

NIH grant NS091201

FAPESP grant 201324660-0

AHA grant 17SDG33410777

Title: Polymerase delta-interacting protein 2 regulates astrocyte activation in ischemic stroke

Authors: *M. S. HERNANDES[1], B. LASSÈGUE[1], L. L. HILENSKI[1], J. ADAMS[2], L. CHENG[3], M. YEPES[3], K. GRIENDLING[1]
[1]Cardiol., [2]Endocrinol., [3]Neurol., Emory Univ., Atlanta, GA

Abstract: Introduction: Polymerase delta-Interacting Protein 2 (Poldip2) is a multifunctional protein that regulates extracellular matrix via its ability to modify secretion of matrix components and alter matrix metalloproteinase activity. The blood-brain-barrier (BBB) is a dynamic structure assembled by endothelial cells, the basal lamina and perivascular astrocytes, raising the possibility that Poldip2 may be involved in maintaining its structure. **Objective:** Investigate the role of Poldip2 in the barrier function of the BBB in the ischemic brain. **Methods:** Transient middle cerebral artery occlusion (tMCAO) was induced in wild type (WT) and Poldip2$^{+/-}$ mice. The volume of the ischemic lesion was measured in TTC-stained sections. BBB breakdown was evaluated by Evans blue dye extravasation. Poldip2 protein expression was evaluated by immunofluorescence and western blotting. RT-PCR was used to measure mRNA levels of cytokines and MMPs. Cultured astrocytes were transfected with Poldip2 siRNA in culture and mRNA levels of cytokines were evaluated. **Results:** Micro-CT and electron microscopy were utilized to investigate the effect of Poldip2 depletion on the brain vasculature. No baseline difference between WT and Polidip2$^{+/-}$ mice was found in vascular volume, vessels per unit area, mean thickness, spacing between vessels, or in the ultrastructure of capillaries. An increase in connectivity (0.05±0.005 vs. 0.08±0.009) was observed (n=10). Poldip2$^{+/-}$ and WT mice displayed comparable infarct sizes following tMCAO (n=6). A decrease in Evans blue dye extravasation was observed in Poldip2$^{+/-}$ mice (25±3 *vs* 6±2µM/g) (n=7). Upregulation of cytokine mRNA following tMCAO was attenuated in Poldip2$^{+/-}$ mice (n=5): MCP-1(253±34 *vs* 83±23AU), IL-6 (134±38 *vs* 38±10AU) and TNFα (39±12 *vs* 12±3AU). MMP-2 (19±3 *vs* 10±1AU), MMP-9 (253±12 *vs* 13±3AU) and TIMP-1 (286±110 *vs* 61±15AU) expression induced by cerebral ischemia was abrogated in Poldip2$^{+/-}$ mice (n=5). Poldip2 protein expression increased in the ischemic brain of WT mice after tMCAO (69±4 *vs* 20±5AU) and was located in perivascular astrocytes (n=4). Poldip2 protein expression was also increased in cultured astrocytes following hypoxia (79±15 *vs* 27±5AU) (n=6), and Poldip2 siRNA prevented cytokine induction under these conditions. Significantly, the protective effect of Poldip2 depletion on increased permeability of the BBB permeability after ischemia was partially reversed by ip injection of TNF-α (5±1 *vs* 24±4µM/g) (n=5).**Conclusions:** Poldip2 contributes to BBB

breakdown after stroke. We propose that Poldip2 is an important regulator of the astrocyte-mediated inflammatory response in cerebral ischemia.

Disclosures: **M.S. Hernandes:** None. **B. Lassègue:** None. **L.L. Hilenski:** None. **J. Adams:** None. **L. Cheng:** None. **M. Yepes:** None. **K. Griendling:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.09/Z32

Topic: C.07. Ischemia

Support: NSF TUES Grant 1245526

Title: Ginkgo biloba down-regulates pro-apoptotic gene expression in an *In vitro* stroke model

Authors: K. M. MCINTYRE[1], F. L. PRENDES[1], K. KARNAS[2], *A. J. ETTINGER[1]
[1]Biol. Sci. and Neurosci. Program, [2]Biol. Sci., Cedar Crest Col., Allentown, PA

Abstract: Ischemic stroke is a leading cause of death and disability in the United States, and current treatment options are limited and time-sensitive. In the brain, cellular apoptosis pathways are active for days following the initial insult and expand the area of damage. *Ginkgo biloba* leaf extract, an herbal remedy long used in traditional medicine, contains several components with antioxidant properties that could block the expansion of apoptosis to neighboring cells. Previous work has demonstrated that the purified extract of *Ginkgo biloba* (EGB761) has neuroprotective effects on cultured cells by blocking apoptosis. This study sought to extend those experiments by examining the effects of an over-the-counter *Ginkgo* supplement that people could easily access as either a prophylactic or an acute treatment. Primary brain and body cell isolates from *Gallus gallus* embryos served as a stroke model in which hydrogen peroxide was used to induce apoptosis, mirroring the cell death that occurs following the initial ischemic event; prophylactic *Ginkgo* treatment of these cell isolates was used to assess the protective properties of this herbal remedy. Cell viability assays using trypan blue exclusion at 0-24 hours post-treatment demonstrated that treated cells had improved survival compared to PBS-treated control cells. Apoptosis-associated gene expression differences were investigated via two independent PCR arrays; these assays showed up-regulation of several anti-apoptotic genes and down-regulation of several pro-apoptotic genes, such as caspases, that are components of the Fas death-inducing signaling complex (DISC), suggesting that *Ginkgo* is acting via this pathway. Western blots will directly examine the expression of some of the protein products of this pathway to determine whether protein level changes are consistent with gene expression changes. As dietary supplements, commercially available *Ginkgo* products are not regulated by the FDA. To test the potential biological activity of these supplements, a spectrophotometric assay was performed to

directly analyze the free-radical scavenging ability of separate containers of over-the-counter *Ginkgo biloba.* A dose-dependent effect of *Ginkgo* was measured, and samples from different lots showed similar radical scavenging. Together, these results merit further study of the use of a commercially available crude herbal supplement to convey protection from cellular apoptosis. Future work will include additional analysis of gene and protein expression and the direct inhibition or activation of the Fas death-inducing pathway to confirm its role in mediating the protective effects of *Ginkgo biloba* extract.

Disclosures: K.M. McIntyre: None. **F.L. Prendes:** None. **K. Karnas:** None. **A.J. Ettinger:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.10/AA1

Topic: C.07. Ischemia

Title: Effect of resveratrol on expression of glucose transporters in astrocytes and neurons subjected to excitotoxicity

Authors: *G. F. GUTIÉRREZ-AGUILAR[1], P. AGUILERA HERNANDEZ[2]
[1]Patología Vascular Cerebral, Inst. Nacional De Neurología Y Neurocirugía, Ciudad DE Mexico, Mexico; [2]Patología Vascular Cerebral, Inst. Nacional De Neurología Y Neurocirugía, Mexico, DF, Mexico

Abstract: Stroke is characterized by a violent diminution of the blood circulation to the brain. Eighty percent of the strokes observed in clinical practice are of the ischemic type, whereas less than 15% are of the hemorrhagic type. Ischemia activates several mechanisms involved in neuronal damage such as excitotoxicity. N-methyl-D-aspartate (NMDA) glutamate receptors are highly permeable to Ca2+ ion channels whose over-activation induces death exclusively in neurons. The extracellular glutamate concentration is finely regulated through the astrocytic Na+-dependent glutamate transporter, the operation of which depends on the Na+/K+ gradient generated across the cell membrane by the Na+/K+ pump. Therefore, the preservation of the transmembrane Na+ gradient signifies an enormous energy cost for the astrocyte. Glucose transporters, GLUT-1 and GLUT-3, are the most abundant in the mammalian brain associated to the high level of expression in astrocytes and neurons, respectively. Under ischemic conditions brain cells regulate GLUT transporters expression in order to compensate its energetic deficiency. Objective. In this work we describe the effect of glutamate treatment on GLUT expression in neuronal and astrocytes cultures. Methods. Neurons were cultured from cerebral cortex of rat embryos (E17-18) an used after 10 DIV. Astrocytes were cultured from neonatal cortex (3 days) and used after 10 - 21 DIV. Cultures were stimulated with 100 μM glutamate for

10 minutes followed by different times recovery (from 1 to 24 h). GLUT expression was evaluated by western blot and immunofluorescence assays. Results and discussion. We observed that glutamate did not alter GLUT3 expression on neurons. On the contrary, astrocytes increase the expression of the “neuronal” glucose transporter (GLUT3), after glutamate neurotoxic stimulation. NMDA receptor activation was not involved on GLUT3 regulation which suggest that the glutamate transporter might be implicated. It has even been described that NF-κB pathway activation promotes transcription and translocation of GLUT3 to the membrane in conditions of oxygen and glucose deprivation which simulates ischemia, although the pathway has not been clearly described. Our results suggest that astrocytes over-express GLUT3 in order to support neuronal energetics under stress conditions such as cerebral ischemia.

Disclosures: G.F. Gutiérrez-Aguilar: None. **P. Aguilera Hernandez:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.11/AA2

Topic: C.07. Ischemia

Support: NIH NS095192

NIH NS099531

VA 101 BX002985

Title: Transient focal ischemia significantly alters the temporal expression profiles of cerebral circular RNAome

Authors: *S. L. MEHTA, G. PANDI, R. VEMUGANTI
Neurolog. Surgery, Univ. of Wisconsin, Madison, WI

Abstract: Circular RNAs (circRNAs) are a novel class of non-coding RNAs (ncRNAs) formed from many protein-coding genes by backsplicing and covalent linking the 3′ end to the 5′ end. Their stability exceeds the linear RNAs due to the absence of well-defined ends. Although their physiologic functions are not yet completely defined, they are thought to control transcription, translation and miRNA levels. We investigated whether stroke changes the circRNAs expression profile in the mouse brain. Male C57BL/6J mice were subjected to transient middle cerebral artery occlusion (MCAO), and circRNA expression profile was evaluated in the penumbral cortex at 6h, 12h, and 24h of reperfusion using circRNA microarrays and real-time PCR. Bioinformatics analysis was performed to identify miRNA binding sites, transcription factor binding and gene ontology of circRNAs altered after focal ischemia. We found that 1,320 circRNAs were expressed at detectable levels mostly from exonic (1,064) regions of the genes in

the cerebral cortex of sham animals. Of those, 283 were altered (>2-fold) at least at one of the reperfusion time points, whereas 16 were altered at all 3-time points of reperfusion after transient MCAO studied compared to sham. Post-ischemic changes in circRNAs identified by microarray analysis were confirmed by real-time PCR. Bioinformatics analysis showed that these 16 circRNAs contain binding sites for many miRNAs. Promoter analysis revealed that the circRNAs altered after stroke might be controlled by a set of transcription factors. The major biological and molecular functions controlled by circRNAs altered after transient MCAO are biological regulation, metabolic process, cell communication, and binding to proteins, ions and nucleic acids. This is a first study that shows that stroke alters the temporal expression of circRNAs with possible functional implication to post-stroke secondary brain damage. Supported by Department of Neurological Surgery-University of Wisconsin, U.S. Department of Veterans Affairs and National Institute of Health.

Disclosures: S.L. Mehta: None. **G. Pandi:** None. **R. Vemuganti:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.12/AA3

Topic: C.07. Ischemia

Support: NIH/NINDS grant NS079153

NIH/NINDS grant NS075035

NIH/NINDS grant NS097000

AHA Fellow to Faculty Award

Title: Putative stroke/vascular risk gene expression in blood of ischemic stroke patients is sexually dimorphic and cause-specific

Authors: *C. J. DYKSTRA-AIELLO, F. R. SHARP, G. C. JICKLING, H. HULL, F. HAMADE, N. SHROFF, X. ZHAN, D. LIU, B. P. ANDER, B. S. STAMOVA
Neurol., Univ. of California, Davis, Davis, CA

Abstract: **Background**. Few stroke/vascular risk factor loci have consistent association with stroke, raising questions about whether risk loci might be specific to stroke cause and/or sex. This study examined stroke/vascular risk factor gene expression in men and women with varying stroke etiologies. **Methods.** Male (n=122) and female (n=123) stroke and vascular risk factor control (VRFC) subjects were matched for race, age, time since stroke and vascular risk factors. Assessment of blood RNA expression for 72 stroke/vascular risk factor genes from whole

transcriptome arrays was completed at gene-, alternative splicing- and exon-levels. Expression of putative risk genes in blood of males and females was also evaluated following cardioembolic (CE), large vessel disease (LVD) and lacunar/small vessel disease (SVD) compared to Vascular Risk Factor Controls (VRFC). **Results**. Statistically significant differential expression (fold change >|1.2|, p-value<0.05, partial time correlation >|0.4|) and alternative splicing (FDR p<0.05) of putative risk genes was sexually dimorphic and stroke cause specific. Post-stroke exon-level expression changes were found in 71 of 72 genes studied, although very few genes were differentially expressed at gene level. These genes included ALDH2, ALOX5AP, F13A1, and IMPA2 in male ischemic stroke (all causes); ITGB3 in female CE; ADD1 in male LVD; F13A1, IMPA2 in male and WNK1 in female SVD. Alternative splicing of GP1BA and ITGA2B was significant in all stroke patients compared to controls in both sexes. Additionally, alternative splicing was found for six genes (ITGB3, LPA, LPAL2, NINJ2, PTGIS, ZFHX3) in males and five genes (F5, MTHFR, PON1, PRKCH, SMARCA4) in females when comparing all stroke to VRFC. Analyses of exon-level expression and differential alternative splicing showed association of many risk loci with a specific stroke cause in either sex. In all stroke patients, genes represented by differentially expressed exons included 24 that were associated with males and 2 (CDC5L/SUPT3H, PRKCH) that were female specific. Six represented genes (ACE, HDAC9, ITGB3, MTHFR, SMARCA4, SORT1) were common to both sexes. Six genes (ADD1, NINJ2, PCSK9, PEMT, SMARCA4, WNK1) in SVD patients were represented by 19 differentially expressed exons with decreased expression in males and increased expression in females. **Conclusion.** Results demonstrate that expression of most of the putative risk genes in blood of IS patients following stroke is both sexually dimorphic and, often, stroke cause specific. Both factors should be included in future stroke treatment trials and genetic association studies.

Disclosures: C.J. Dykstra-Aiello: None. **F.R. Sharp:** None. **G.C. Jickling:** None. **H. Hull:** None. **F. Hamade:** None. **N. Shroff:** None. **X. Zhan:** None. **D. Liu:** None. **B.P. Ander:** None. **B.S. Stamova:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.13/AA4

Topic: C.07. Ischemia

Support: SHU BioResearch Fund

Title: Alteration of human neuronal kappa opioid receptor expression under hypoxic mimic condition

Authors: J. BABCOCK, A. HERRERA, G. CORICOR, C. KARCH, A. LIU, A. RIVERA-GINES, *J. L. KO
Dept. of Biol. Sci., Seton Hall Univ., South Orange, NJ

Abstract: Cellular adaptation to stressors, like hypoxia, is essential for neuronal survival. Some cells in a hypoxic environment developed adaptive changes allowing them to survive while others die as a result of the decrease in oxygen. Using human neuronal NMB cells treated with the hypoxic mimetic DFO, a hypoxic mimic cell model system was generated. A reduction of cell viability was detected. The cells surviving DFO exposure exhibited an increase in expression of the kappa-opioid receptor (KOR) and hypoxia inducible factor-1 alfa (HIF-1a). Therefore, the relationship between HIF-1a and KOR was investigated using RT-PCR, Western blot, luciferase reporter assay, mutagenesis, siRNA, and receptor-ligand binding assays. Neurons surviving DFO exposure exhibited an increase in HIF-1a expression overall, and its level in the nucleus was also increased. The surviving neurons also expressed an increase of KOR mRNA level in the time dependent manner as comparison to the control sample. While knockdown of HIF-1a by siRNA inhibited the increase of endogenous HIF-1a, it also reduced DFO-induced KOR expression. The HIF-1a siRNA treatment did not significantly affect the endogenous level of HIF-2a. To further examine the role HIF-1a in hKOR regulation, analysis of HREs of KOR gene was performed. Results revealed four potential HIF response elements (HIFA-D). Two of these HREs (HIFC and HIFD) synergistically mediated the DFO response using the reporter assay, also exhibiting a dose-dependent manner. Upon mutation of these two elements, DFO-induced effect was completely abolished. The CD1 plasmid, containing HIFC and HIFD with an 11 bp spacer, yielded a greater increase in promoter activity compared to that of CD17 plasmid (with a 17 bp spacer), suggesting that a head-tail arrangement of HREs synergistically enhanced promoter activity. Taken together, the alteration of human KOR expression under DFO treatment was mediated by HIF-1 in neuronal cells. Further studies are to determine the impacts of hKOR expression under hypoxic mimic condition.

Disclosures: J. Babcock: None. **A. Herrera:** None. **G. Coricor:** None. **C. Karch:** None. **A. Liu:** None. **A. Rivera-Gines:** None. **J.L. Ko:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.14/AA5

Topic: C.07. Ischemia

Support: Research supported by NIH grant 5R01MH086638

Title: Multiscale computer modeling of penumbral zones in brain ischemia

Authors: *A. SEIDENSTEIN[1], A. NEWTON[2], R. A. MACDOUGAL[3], W. W. LYTTON[4]
[1]CBE, New York University- Tandon Sch. of Engin., Brooklyn, NY; [2]Neurosci., [3]Dept. of Neurosci., Yale Univ., New Haven, CT; [4]Dept. Physiol. & Pharmacol., SUNY Downstate Med. Ctr., Brooklyn, NY

Abstract: On a cellular level, molecular cascades produce damage that can lead to apoptosis, necrosis or necroptosis. Damage propagates due to a combination of extracellular diffusion and synaptic connectivity. Temporally, we are interested in aberrant spiking activity over milliseconds up to the progression of ischemia extending over a period of hours. Spatial scales range from subcellular changes in ion concentrations to large regions of brain tissue. Tissue affected by ischemic stroke is divided into three regions; 1. the core where cells suffer irreparable damage and death, 2. a penumbra where cells may recover with reperfusion, 3. a further region of edema where spontaneous recovery is expected.We extended the NEURON reaction-diffusion modules (NRxD) to include the extracellular space (ECS), providing a platform for modeling ischemia, with the penumbra cells and embedded in its extracellular environment. Here, we explored cellular failure initiated by ATP depletion at a central location. Ischemia impedes ATP production which results in a failure of the Na+/K+-ATPase pump and a rise in extracellular K+. Once extracellular K+ exceeds a threshold it will cause neurons to depolarize, further increasing extracellular K+. The cell network was created using NetPyNE in order to model the spreading depression seen in ischemic stroke by coupling a detailed biophysical model of cortical pyramidal neurons equipped with Na+/K+-ATPase pumps with reaction-diffusion of ions in the ECS.Multiscale modeling of ischemia combines the dynamics of rapid synaptic connectivity and slower extracellular diffusion of ions and toxic products of cell death, giving varying effects of core on the penumbra. Different penumbral subzones will produce differing intracellular dynamics biasing the cell to either cell death or potential recovery. The tools presented in this model allow for more efficacious experimentation and understanding of potential mechanisms to prevent permanent penumbra cell death.

Disclosures: A. Seidenstein: None. **A. Newton:** None. **R.A. MacDougal:** None. **W.W. Lytton:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.15/AA6

Topic: C.07. Ischemia

Title: Identification the key sites of S2-S3 loop involved in TRPM2 gating mechanism

Authors: *Y. LUO, W. YANG
Inst. of Neurosci., Zhejiang province, China

Abstract: The melastatin-related transient receptor potential channel TRPM2 is a plasma membrane Ca^{2+}-permeable cation channel and it is mainly activated by intracellular adenosine diphosphoribose (ADPR) with the existence of Ca^{2+}. However, the mechanism of ADPR coordinated with calcium to activate TRPM2 channel remains unknown. By employing Rosetta software, we predicted the structure modeling of TRPM2 transmembrane domain based on TRPV1 structure. Interestingly, our model shows S2-S3 loop, the second longest loop between transmembrane segments, which contains α-helix and closes to the S4-S5 linker. By performing sequencing alignment across TRPM1-8 family, we found several amino acids are highly conserved including E842. In addition, several amino acids are conserved in TRPM2, TRPM4 and TRPM5, such as E843 and so on. It is well known that TRPM2, TRPM4 and TRPM5 are activated by calcium instead of other TRPM channels, which suggests there are some key sites to determine the calcium activated these channels. Here, we tested the hTRPM2 and its mutants currents in HEK293 cells by using 500μM ADPR, 50mM Ca^{2+},500μM ADPR+100μM Ca^{2+} conditions, respectively. Our results indicated that several amino residues (from L836 to P851) are critical for TRPM2 activation. Strikingly, E842A was no response to ADPR but had a small response to Ca^{2+}, while E843K was no response to Ca^{2+} but had a small response to ADPR, which suggests E842A and E843K separated the process of ADPR and Ca^{2+} activated TRPM2. Since E843 residue only exists in TRPM2/4/5 channels, our results strongly suggests that E843 is responsible for Ca^{2+} but not ADPR gating TRPM2 channel. In summary, our findings for first time identified key residues to separate the TRPM2 activation by ADPR or Ca^{2+}, which will be used to clarify the contribution of Ca^{2+} or ADPR alone in TRPM2 mediated neuron diseases such as ischemia injury and Alzheimer's disease.
Keywords:TRPM2 ,S2-S3 loop, ADPR,Ca^{2+} ,channel gating

Disclosures: **Y. Luo:** None. **W. yang:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.16/AA7

Topic: C.07. Ischemia

Support: Kakenhi 17K08512

Takeda Science Foundation

SENSHIN Medical Research Foundation

Title: Netrin-5 is upregulated in the peri-infarct region after middle cerebral artery occlusion

Authors: *S. YAMAGISHI[1], M. TAKARADA[2], M. SAWADA[3], K. SAWAMOTO[3], O. HORI[2], K. SATO[1]
[1]Organ and Tissue Anat., Hamamatsu Univ. Sch. of Med., Hamamatsu-Shi, Japan; [2]Kanazawa Univ., Kanazawa, Japan; [3]Nagoya City Univ. Grad. Sch. of Med. Sci., Nagoya, Aichi, Japan

Abstract: Mammalian netrin family proteins are involved in targeting of axons, neuronal migration, and angiogenesis and act as repulsive and attractive guidance molecules. Netrin-5 is a new member of the netrin family with homology to the C345C domain of netrin-1. Strong netrin-5 expression was observed in the olfactory bulb (OB), rostral migrate stream (RMS), the subventricular zone (SVZ), and the subgranular zone (SGZ) of the dentate gyrus in the hippocampus, where neurogenesis occurs in the adult brain. In the SVZ and RMS, netrin-5 expression was observed in Mash1-positive transit-amplifying cells and in Doublecortin (DCX)-positive neuroblasts, but not in GFAP-positive astrocytes. In the OB, netrin-5 expression was maintained in neuroblasts, but its level was decreased in NeuN-positive mature neurons. In the hippocampal SGZ, netrin-5 was observed in Mash1-positive cells and in DCX-positive neuroblasts, but not in GFAP-positive astrocytes, suggesting that netrin-5 expression occurs from type 2a to type 3 cells. Next, we generated ischemia mouse model with middle cerebral artery occlusion. After 3 days from the stroke, netrin-5 was highly upregulated in the infarct region. Netrin-5 positive cells were expressing both NG2 and Iba1, suggesting they are BINCS (Brain Iba1+ /NG2+ Cells) derived from bone marrow and beneficial. These data suggest that netrin-5 is involved in neurogenesis and phagocytosis in the peri-infarct region of the cortex.

Disclosures: S. Yamagishi: None. **M. Takarada:** None. **M. Sawada:** None. **K. Sawamoto:** None. **O. Hori:** None. **K. Sato:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.17/AA8

Topic: C.07. Ischemia

Support: Neuroscience Institute, JFK Medical Center

Title: Exploring the genome’s dark matter for novel therapeutic targets against ischemic stroke

Authors: *S. BHATTARAI, F. PONTARELLI, A. DHARAP
Neurosci. Inst., JFK Med. Ctr., Edison, NJ

Abstract: Long noncoding RNAs (lncRNAs) derived from the ‘dark matter’ of the genome play major roles in regulating gene expression in mammals, but their expression and functions after ischemic stroke are poorly understood. Using a mouse model of transient focal ischemia, we

applied RNA-sequencing to evaluate for the first time, the unbiased, genome-wide expression of lncRNAs as a function of reperfusion time in the cerebral cortex. After a 1h middle cerebral artery occlusion in adult male mice, ipsilateral cortices were harvested at 6, 12 or 24h of reperfusion. RNA was isolated and used for Illumina deep sequencing. The reads were mapped to the mouse genome (GRCm38), annotated and novel transcript isoforms were identified, and differential expression between the experimental groups was quantified. The data was validated using real-time PCR. Our results showed that while the baseline expression of lncRNAs in the healthy cortex was low, many of them were highly induced after stroke. In addition, hundreds of genomic loci that are not transcribed in the healthy cortex were robustly induced in the post-ischemic cortex, yielding ischemia-specific lncRNAs. Very few of the altered lncRNAs were previously annotated. We identified 259 transcript isoforms at 6h, 378 isoforms at 12h, and 217 isoforms at 24h of reperfusion that were differentially expressed versus sham controls. Of these, 212, 322 and 171 isoforms at 6, 12 and 24h of reperfusion, respectively, were novel lncRNAs. Overall, this work shows that transient focal ischemia induces widespread changes in the expression of lncRNAs in the mouse cortex with distinct reperfusion time-point-dependent expression characteristics that may underlie progression of the ischemic pathophysiology. The detection of hundreds of novel lncRNAs marks the discovery of new disease-related genomic regions in the adult post-stroke cortex.

Disclosures: **S. Bhattarai:** None. **F. Pontarelli:** None. **A. Dharap:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.18/AA9

Topic: C.07. Ischemia

Support: PRIN Grant 2015E8EMCM_006

PRIN Grant 2015E8EMCM_002

Title: The selective antagonism of adenosine A_{2b} receptors prevents synaptic and neuronal damage induced by oxygen and glucose deprivation in CA1 rat hippocampus

Authors: F. UGOLINI, D. LANA, I. FUSCO, E. COPPI, I. DETTORI, L. GAVIANO, F. PEDATA, G. PEPEU, A. PUGLIESE, *M. GIOVANNINI
Univ., Firenze, Italy

Abstract: Cerebral ischemia results from severe reduction of cerebral blood flow after cardiac arrest, occlusion of vessels supplying nervous tissues, or prolonged systemic hypotension. Each year about 700,000 people suffer new or recurrent stroke, a major cause of long-term disability,

and the third most common cause of death in Western countries. The availability of drugs able to counteract stroke induced neurodegeneration is still an unmet need. Extracellular concentrations of adenosine significantly increase during ischemia reaching the μmolar range, activating all adenosine receptor subtypes: A_1, A_{2A}, A_{2B}, and A_3. No data on the involvement of A_{2B} receptor in cerebral ischemia are so far available. In this work we investigated the role of the A_{2B} receptor during oxygen and glucose deprivation (OGD, 7 min), a model of cerebral ischemia, in CA1 of rat hippocampus in vitro. A selective antagonist for A_{2B} receptor, PSB 603, was used to investigate the receptor putative involvement in the mechanisms of ischemic-mediated neuronal death. In order to better characterize the OGD-induced cell injury and pharmacological protection, if any, we conducted extracellular recordings of CA1 field excitatory post-synaptic potentials (fEPSPs); the extent of damage on neurons and glia was assessed by immunohistochemistry. OGD induced anoxic depolarization (AD), an unambiguous sign of neuronal damage, in all hippocampal slices tested (n=38), and completely abolished fEPSPs that did not recover after return to normoxic condition. The selective antagonist of A_{2B} receptor PSB 603 (50 nM, n=9) significantly delayed the appearance of AD and recovered fEPSPs amplitude. The extent of CA1 pyramidal neurons injury was assessed 1 hour after the end of OGD by immunofluorescence for NeuN. OGD caused substantial damage on CA1 pyramidal neurons, as determined by the significant increase of pyknotic nuclei (+696%, n=6), and of degenerating, anucleated neurons (+1400%, n=6). These effects were completely blocked by the antagonist PSB 603 (50 nM, n=5). OGD did not affect astrocytes or microglia, at least 1 h after the end of OGD. These results are the first to show that selective antagonism on the adenosine A_{2B} receptor delays the occurrence of AD and improves neuronal survival following severe OGD in the CA1 hippocampus, as demonstrated by the significant recovery of an otherwise disrupted neurotransmission and by the significant attenuation of neuronal damage induced by OGD. The selective antagonists for the A_{2B} adenosine receptor subtype may thus represent a new class of neuroprotective drugs.

Disclosures: **F. Ugolini:** None. **D. Lana:** None. **I. Fusco:** None. **E. Coppi:** None. **I. Dettori:** None. **L. Gaviano:** None. **F. Pedata:** None. **G. Pepeu:** None. **A. Pugliese:** None. **M. Giovannini:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.19/AA10

Topic: C.07. Ischemia

Support: Dept. of Veterans Affairs

NIH (NS081149)

Title: Cofilin-actin rod formation in brain ischemia

Authors: *S. WON[1], A. M. MINNELLA[1], C. EUN[1], E. ROME[1], P. S. HERSON[2], F. DABERTRAND[3], J. R. BAMBURG[4], R. A. SWANSON[1]
[1]Neurol, UCSF and SFVAMC, San Francisco, CA; [2]Anesthesiol., UC Denver, Aurora, CO; [3]Pharmacol., Univ. of Vermont, Burlington, VT; [4]Colorado State Univ., Fort Collins, CO

Abstract: Cofilin is a member of actin depolymerizing factor (ADF)/cofilin family, and plays a key role in the regulation of actin turnover (treadmilling). In neurons, actin turnover is involved in vesicle and organelle trafficking, and in the formation and maintenance of dendritic spines. Under stress conditions such as low ATP and oxidative stress, cofilin and actin form reversible rod-shaped aggregates. These may function to stop actin turnover and vesicular release, thereby preserving ATP consumed by these processes. However, the sustained presence of cofilin-actin rods may lead to spine collapse and degeneration of distal processes. It is unknown whether cofilin-actin rod formation contributes to stroke pathophysiology. As an initial step, we characterized cofilin-actin rod formation in cell culture and in five mouse models of brain ischemia. In 16-day old cortical neuron cultures, both glutamate exposure and oxygen glucose deprivation induced cofilin-actin rod formation in the neuronal processes. This was suppressed by inhibition of NADPH oxidase-2 (which produces superoxide). Cofilin-actin rod formation in vivo was assessed after transient forebrain ischemia, transient middle cerebral artery occlusion, cardiac arrest and cardiopulmonary resuscitation (CA/CPR), permanent photothrombotic ischemia, and in a mouse model of CADASIL (cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy). We observed neuronal cofilin-actin rod formation in each of these models. Rod formation was most evident in and adjacent to the ischemic territories, and sometimes extended into neighboring white matter tracts. In the transient ischemia models, rod formation was most evident shortly after reperfusion, but was still evident 24 hours later. Ongoing studies are evaluating the effects of cofilin-actin rod formation on subsequent neurite survival.

Disclosures: S. Won: None. **A.M. Minnella:** None. **C. Eun:** None. **E. Rome:** None. **P.S. Herson:** None. **F. Dabertrand:** None. **J.R. Bamburg:** None. **R.A. Swanson:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.20/AA11

Topic: C.07. Ischemia

Support: U.S. Veterans Affairs Merit grant BX000917

Title: NAD^+ precursor protects against ischemic brain injury by promoting mitochondrial fusion

Authors: N. KLIMOVA[1], A. LONG[2], *T. KRISTIAN[3]
[1]Anesthesiol., Univ. of Maryland, Baltimore, Baltimore, MD; [2]Anesthesiol., Univ. of Maryland, Baltimore, MD; [3]Dept Anesthesiol, Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: Ischemic insult depletes pools of NAD^+, an essential factor in mitochondrial and cellular metabolism, leading to bioenergetics failure and cell death. NAD^+ levels can be replenished by the administration of nicotinamide mononucleotide (NMN), which serves as a biochemical precursor for NAD^+ synthesis. To assess the effect of NMN administration on ischemia-induced mitochondrial dynamics we used transgenic mouse model with mitochondria targeted expression of neuron specific enhanced yellow fluorescent protein (eYFP). The animals were subjected to transient forebrain ischemia and NMN was administered 30 minutes after the start of reperfusion at a dose of about 60 mg/kg. CA1 neuronal morphology was analyzed using cresyl violet staining at 6 days of recovery post ischemia. Alterations in mitochondrial fission and fusion dynamics were analyzed in post-ischemic animals treated with vehicle or NMN at 2, 4, and 24 hours of recovery. Fusion and fission protein levels were also determined by western blots. NMN treatment decreased neuronal death in the CA1 sector of the hippocampus and improved neurologic outcome. NMN reversed the excessive mitochondrial fragmentation in CA1 neurons that was permanent in vehicle-treated ischemic groups. Thus, NMN administration promoted normal mitochondrial morphology at 24 hours after the ischemic insult. Western blot analysis showed that NMN administration reversed ischemia-induced increase in P-Drp1(S616) to Drp1 ratio as well as normalized the increase in acetylation of mitochondrial proteins observed after ischemic insult. This effect could be mediated through increased activity of NAD^+ - dependent deacetylases, particularly the mitochondrial localized SIRT3, due to the replenished NAD^+ levels. Our data thus suggest that maintaining normal mitochondrial morphology is essential for cell bioenergetics metabolism and survival. Furthermore, the mechanisms of fission and fusion regulation could represent new therapeutic targets for neurobiological diseases.

Disclosures: N. Klimova: None. **A. Long:** None. **T. Kristian:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.21/AA12

Topic: C.07. Ischemia

Support: NIH R01 NS085019

Title: Matrilin-2: A pro-growth extracellular matrix protein in the post-stroke brain

Authors: *S. P. BRIDGES, M. MACHNICKI, S. T. CARMICHAEL
Dept. of Neurol., Univ. of California Los Angeles, Los Angeles, CA

Abstract: Ischemic brain injury causes many changes in the cells and environment of the central nervous system, leading to an incomplete recovery process and lasting disability due to failure to regenerate preexisting circuits. Changes in the extracellular matrix of the post-stroke brain have long been speculated as a primary cause for this halted recovery, with a great deal of focus being placed on a growth inhibitory environment following CNS insult. Transcriptional profiling of neurons that sprout after stroke shows upregulation of several extracellular matrix proteins that may be beneficial to the endogenous repair process; one such protein is matrilin-2. Matrilin-2 is a cartilage associated protein that is important in peripheral nerve regeneration following injury, but its function in the CNS is unknown. In our study, we assessed the pro-growth effect of matrilin-2, using cultured primary cortical neurons as well as in the CNS post stroke. Matrilin-2 was found to enhance axonal outgrowth in a dose dependent manner in post-natal day 3 cortical neurons, and was capable of overcoming a growth inhibitory environment *in vitro*. To assess the ability of matrilin-2 to promote enhanced axonal sprouting post-stroke, adult C57Bl/6 mice were subjected to photothrombotic motor cortex stroke and lentiviral overexpression of matrilin-2 in the peri-infarct region. Axonal sprouting relative to stroke location was assessed via BDA tracing of axons from the peri-infarct region. Our results begin to explore the pro-regenerative ECM environment in the post-stroke brain, and its role in functional recovery after stroke.

Disclosures: S.P. Bridges: None. **M. Machnicki:** None. **S.T. Carmichael:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.22/AA13

Topic: C.07. Ischemia

Support: FONDECYT 1140477 (FN)

11140405 (KS)

CMA BIO BIO, CONICYT ECM-12

Title: Vitamin C and neuronal oxidative stress alter mitochondrial and cellular size, triggering neuronal death

Authors: *L. E. FERRADA[1], K. A. SALAZAR[3], F. J. NUALART[2]
[1]Univ. de Concepción, Concepcion, Chile; [2]Univ. de Concepción, Concepción, Chile; [3]Univ. De Concepción, Concepcion, Chile

Abstract: Vitamin C is the principal antioxidant in the central nervous system and is found in two states: reduced ascorbic acid (AA) and oxidized dehydroascorbic acid (DHA). In the presence of oxidative damage, AA acts as an antioxidant, oxidizing to DHA. Whereas AA is

incorporated by neurons through the sodium-dependent vitamin C transporter (SVCT2), DHA diffuses into nerves by glucose transporters (GLUTs). Our laboratory has shown that accumulation of DHA induces neuronal death in oxidative stress, but the cell death pathways are unknown. Here, we analyzed cellular and mitochondrial morphology, because morphological alterations and mitochondrial fission/fusion processes are associated with cell death. The Neuro2a cell line and rat cortical neurons were supplemented with AA and thereafter treated with H_2O_2. Cell viability was quantified using the XTT method. Cellular distribution of SVCT2 and GLUT1 were analyzed by SIM super-resolution microscopy. Mitochondrial and cellular size was analyzed using the Bounding-Box tool. Oxidation of vitamin C generates neuronal death in 50% of the cells. Prior to neuronal death, GLUT1 distribution was altered from the plasma membrane to the perinuclear zone and SVCT2 increased in the plasma membrane. Oxidation of vitamin C decreased mitochondrial size and volume on average by 0.3 µm and 0.2 μm^3, respectively. Furthermore, we noted a decrease in mitochondrial activity (MitoTracker, red) with 4D live cell experiments following permeabilization of the plasma membrane and incorporation of phalloidin (green). In conclusion, intracellular DHA generation induces neuronal death, which involves the redistribution of SVCT2 and GLUT1. Mitochondrial size, volume, and activity also decrease during vitamin C oxidation.

Disclosures: L.E. Ferrada: None. **K.A. Salazar:** None. **F.J. Nualart:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.23/AA14

Topic: C.07. Ischemia

Support: NIH Grant NS073832-01A1 (AZ)

NIH Grant 5U54NS083932 (NI)

NIH Grant S21MD000101 (MSM)

NIH Grant G12-RR03034 (RCMI) (MSM)

Title: Epigenetic regulation of ion channels in stroke, the acts of PcG and TrxG proteins

Authors: *A. ZHOU[1], L. HERNANDEZ[1], P. SHARMA[1], T. YANG[1], R. P. SIMON[2]
[1]Morehouse Sch. of Med., Atlanta, GA; [2]Morehouse Med. Sch., Atlanta, GA

Abstract: Ion channels play pivotal roles in the pathology of ischemic stroke. Little is known if and how different ion channels are synergistically regulated in brain ischemia. Polycomb group (PcG) and Trithorax group (TrxG) proteins are epigenetic gene repressors and activators,

respectively, that regulate the same genes via specific histone modifications. Previously, we reported increased levels of several PcG proteins in ischemic-tolerant mouse brains (Stapels et al., Sci Signal. 2010 Mar 2;3(111):ra15). We showed neuroprotective roles of PcG proteins against ischemic injury, as well as initial experimental evidence supporting a suppressive role of PcG proteins on whole cell potassium currents. Our recent, unpublished data (presented elsewhere) show that PcG proteins also down regulate sodium currents in neuronal cells. TrxG proteins, on the other hands, appear exert opposite effects on neuronal ischemic injury and ion channel activity. These implicate the play of a previously undescribed, PcG/TrxG protein-mediated epigenetic mechanism in the regulation of ion channels in neuronal ischemia.
The objective of this work was to characterize the expressional changes in ion channel genes in ischemia, with a primary focus on potassium and sodium channel genes, and to determine if their expression can be regulated by PcG or TrxG proteins. In mice in vivo, focal cerebral ischemia was induced by middle cerebral artery occlusion. In cultured neuronal cells in vitro, simulated ischemia was modeled by oxygen-glucose deprivation. Three ischemic conditions were included: injurious (prolonged ischemia), preconditioning (short ischemia to induce tolerance), and tolerant ("injurious" ischemia preceded by preconditioning ischemia), plus control. The effects of over- or under-expression of a PcG or TrxG protein on gene expression were examined in normoxic cell cultures. Expression levels of a panel of 84 neuronal ion channel genes (including 42 potassium and 9 sodium channel genes) were determined by qPCR mini-array, and compared across ischemic conditions, or between cells with or without PcG or TrxG manipulation. We found that levels of many potassium channel genes were decreased under ischemic-tolerant conditions, notably members of delayed rectifying and modifier subunit families. Knocking down a PcG protein or inhibiting its histone modification activity resulted in increased expression of a large number of potassium channel genes. Similar changes were also observed for several sodium channel genes. Overall, current results support a role of PcG proteins as broad-base regulators of ion channel genes. Ongoing experiments focus on the roles of TrxG proteins.

Disclosures: **A. Zhou:** None. **L. Hernandez:** None. **P. Sharma:** None. **T. Yang:** None. **R.P. Simon:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.24/AA15

Topic: C.07. Ischemia

Support: Grants-in-Aid and by special coordination funds from Grants-in-Aid for Scientific Research (C) (17K01882) from the Ministry of Education, Culture, Sports, Science and Technology of Japan

Title: Suppressive effect of hypothalamic orexin-A-mediated vagus nerve activation on the cerebral ischemia-induced elevation of hepatic inflammatory cytokines

Authors: *S. HARADA, S. TOKUYAMA
Kobe Gakuin Univ., Kobe, Japan

Abstract: We have previously found that the development of post-ischemic glucose intolerance is one of the triggers of ischemic neuronal damage. Orexin-A (a neuropeptide in the hypothalamus) plays an important role in many physiological functions, including the regulation of glucose metabolism. Activation of vagus nerve exerts orexin-A (OXA)-mediated suppression of post-ischemic glucose intolerance and cerebral ischemic neuronal damage. Cerebral ischemia induces hepatic inflammatory factors and contributes to the development of hepatic insulin resistance by activating sympathetic nerves. However, it is not enough to understand whether OXA regulates these phenomena through activation of vagus nerve. In this study, we demonstrated that the involvement of OXA-induced activation of vagus nerve in the induction of hepatic inflammatory factors by cerebral ischemia.
Male ddY mice were subjected to middle cerebral artery occlusion (MCAO) for 2 h. The hepatic vagotomy mice created to selectively transect at the hepatic branch vagus nerve. Neuronal damage was estimated by histological and behavioral analyses. Expression of each protein levels was analyzed by western blot and immunofluorescence staining.
Intrahypothalamic orexin-A (5 pmol/mouse) administration significantly suppressed the development of post-ischemic glucose intolerance and neuronal damage on day 1 and 3, respectively after MCAO. The MCAO-induced decrease in hepatic insulin receptors and increase in hepatic gluconeogenic enzymes were suppressed by OXA administration. These effects were canceled by *N*-butylscopolamine and hepatic vagotomy. MCAO-induced increases in hepatic tumor necrosis factor-α, and interleukin-1β on day 1 after MCAO were reversed by OXA administration. These effects were abolished by *N*-butylscopolamine or hepatic vagotomy. These results suggest that the orexin-A and vagus nerve may play an important role in the regulation of post-ischemic elevated hepatic inflammatory factors.

Disclosures: **S. Harada:** None. **S. Tokuyama:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.25/AA16

Topic: C.07. Ischemia

Support: Heart and Stroke Foundation of Canada

Saudi Arabia - Ministry of Higher Education

Title: Survival of pyramidal neurons within the ischemic core of 12-hour post-MCAo mice

Authors: *R. H. MEHDER, D. PETRIN, P. J. GAGOLEWICZ, B. M. BENNETT, A. Y. JIN, R. D. ANDREW
Biomed. & Mol. Sci., Queen`S Univ., Kingston, ON, Canada

Abstract: During the acute ischemia of focal stroke, neurons in the higher brain immediately depolarize, swell and lose synaptic input as their dendrites contort and form interconnected beads. At some point the neurons either partially recover or initiate necrosis but the time course is not well described. Here we examined neuronal morphology and electrophysiology at two time points following intraluminal left middle cerebral artery occlusion (MCAo) induced for 30 minutes in male C57/BL6 mice (20-25g). We not only gauged deterioration but also looked for surviving neurons within the ischemic core. The first time-point examined was immediately post-MCAo where there was ~10 minute reperfusion before immersion fixation of the brain. Golgi-Cox stained pyramidal neurons in the future core region were surprisingly unaltered based on morphometric analysis of dendritic branching, length and spine numbers (n=3 mice). Neuronal cell bodies were of comparable size and shape in both non-ischemic and ischemic hemispheres. This is in sharp contrast to the ischemic hemisphere 12h post-MCAo (n=3 mice) where most pyramidal and all striatal neurons were severely dysmorphic. Every principle cell in striatum was shrunken with retracted and beaded dendrites. The pyramidal cell response was more variable, displaying regions of uniformly swollen cell bodies but maintaining a pyramidal shape. Their dendrites lacked spines and were beaded, typical of ischemic neurons in the early phase of deterioration. More commonly, the remaining neocortical neurons were necrotic i.e., shrunken with retracted dendrites lacking spines. This contrasted with neocortical gray immediately outside the MCAo infarct region which appeared normal. Surprisingly, some core pyramidal neurons survived 12h post-MCAo, displaying spinous dendrites and normal cell body size/shape despite being surrounded by swollen or necrotic neurons. These rare cells could be visualized with contrast optics and recorded under whole-cell current clamp in live brain slices. The mean values for the fast afterhyperpolarization, whole cell input resistance, resting membrane potential, as well as action potential threshold, amplitude and width were not significantly different between these 12h `healthy` neurons in the ischemic hemisphere (n=5 cells) and unstressed neurons in the non-ischemic hemisphere (n=6). How long these resilient neurons continue to survive beyond 12h and how they selectively survive are two questions we are currently examining.

Disclosures: R.H. Mehder: None. **D. Petrin:** None. **P.J. Gagolewicz:** None. **B.M. Bennett:** None. **A.Y. Jin:** None. **R.D. Andrew:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.26/AA17

Topic: C.07. Ischemia

Support: Health and Medical Research Fund 03142256

Title: Autophagy in diabetic retinal ischemia/reperfusion injury

Authors: *A. C. LO, L. H. C. TANG, A. K. W. LAI, F. K. C. FUNG
Dept. of Ophthalmology, The Univ. of Hong Kong, Hong Kong, Hong Kong

Abstract: Purpose
Retinal ischemia/reperfusion (I/R) injury is a common cause of retinal cell death. Despite increasing interest in autophagy modulation as a therapeutic strategy, the effect of retinal I/R injury on autophagy (up or downregulation) remains controversial. We investigated the effect of retinal I/R injury on autophagy using both an *in vivo* model and an *in vitro* model.
Methods
In vivo: A middle cerebral artery occlusion (MCAO) stroke model was used to induce unilateral retinal I/R injury in wild-type and Akita (type I diabetic) mice. Ischemia was maintained for two hours, followed by 2 or 22 hours of reperfusion. Autophagy markers, microtubule-associated light chain protein (LC3) and lysosomal associated membrane protein (LAMP1), were assessed via immunohistochemical staining on retinal sections.
In vitro: R28 cells (a retinal precursor cell line) were cultured in low glucose DMEM medium. One group of cells was incubated with additional glucose to mimic hyperglycemia. Hypoxia was chemically induced using cobalt (II) chloride for 24 hours. Cell viability and autophagy was assessed via the MTS assay and the detection of LC3 by immunoblotting, respectively.
Results
In vivo: After 2 hours of reperfusion, LC3 was significantly upregulated in the ganglion cell layer (GCL) in MCAO-injured Akita retinae as compared with sham-treated Akita retinae. After 22 hours of reperfusion, LAMP1 immunoreactivity was increased in the GCL and inner nuclear layer of MCAO-injured retinae as compared with their sham-treated counterparts.
In vitro: Hypoxic cells had lower cell viability than untreated cells. Hypoxia also resulted in significant LC3 upregulation.
Conclusion
Retinal I/R injury and $CoCl_2$-induced hypoxia induces autophagy activation in mouse retinae and R28 cells, respectively. Both our *in vivo* and *in vitro* results suggest that retinal I/R injury-induced autophagy may be exacerbated by elevated glucose levels.

Disclosures: A.C. Lo: None. **L.H.C. Tang:** None. **A.K.W. Lai:** None. **F.K.C. Fung:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.27/AA18

Topic: C.07. Ischemia

Support: NIH Grant NS51288

NIH Grant T32 HL007736

Title: Targeting the vulnerable phase of SD with ketamine does not prevent plasticity mechanisms

Authors: *K. M. REINHART, R. J. OLIVER, G. PERALES, N. I. PERRONE-BIZZOZERO, C. W. SHUTTLEWORTH
Univ. of New Mexico HSC, Albuquerque, NM

Abstract: Spreading depolarizations (SD) are slowly propagating waves of neuronal and glial depolarization that occur in stroke brain. SD causes irrecoverable injury to metabolically compromised tissues by over-excitation of NMDA receptors. However, SD may also promote NMDA receptor-dependent synaptic plasticity and protective preconditioning in healthy tissues surrounding infarcts. We tested whether low concentrations of the NMDA receptor antagonist ketamine can prevent NMDA receptor-dependent injury, without preventing potential beneficial effects of SD. Acute cortico-hippocampal brain slices were prepared from C57Bl/6 mice. SD was initiated by KCl microinjection in CA1 and we monitored extracellular DC potential and evoked excitatory postsynaptic potentials (fEPSPs). BDNF mRNA was assessed by rt-qPCR. We first established a concentration of ketamine (30µM) that did not prevent SD initiation or propagation, but which protected tissues against deleterious effects of SD in vulnerable tissues. DC shifts of SD were reduced, recovery of intracellular Ca^{2+} was accelerated, and tissues recovered the ability to generate repetitive SDs. A post-hoc analysis of fEPSP amplitude revealed that fEPSP potentiation after recovery from SD was not prevented by ketamine ($P = 0.135$; n=11, 8, untreated and ketamine, respectively). In a subset of experiments, total BDNF (BDNF-Pan) and activity-dependent BDNF (BDNF-L) mRNA from hippocampus was assessed 15 minutes post SD. When SD was blocked with high concentrations of ketamine (300µM), both total BDNF and BDNF-L expression were reduced (by ~27% and ~50%, respectively), compared to SD in control conditions. In contrast, when SD propagated in the presence of ketamine (30µM) both BDNF-Pan and BDNF-L mRNA expression were elevated above control levels (by ~73% and ~52%, respectively). These findings suggest that low ketamine concentrations can protect against damaging effects of SD while permitting propagation into peri-infarct regions. Such SDs could potentially contribute to protective preconditioning and support recovery and repair.

Disclosures: **K.M. Reinhart:** None. **R.J. Oliver:** None. **G. Perales:** None. **N.I. Perrone-Bizzozero:** None. **C.W. Shuttleworth:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.28/AA19

Topic: C.07. Ischemia

Title: Pathologic roles of proteolytic enzymes associated with receptor processing

Authors: *M. YAMADA, H. HAYASHI, N. MATSUSHIMA, M. OHATA, B. YUAN, N. TAKAGI
Applied Biochem., Tokyo Univ. of Pharm. and Life Sci., Hachioji Tokyo, Japan

Abstract: Ischemic brain damage is associated with various causes such as glutamate excitotoxicity, oxidative stress and trophic factor deficiency. An excess amount of extracellular glutamate induced by ischemia is a major factor to induce intracellular Ca^{2+} overload via *N*-methyl-D-aspartate (NMDA) receptor and then causes neuronal cell death. Although it has been shown that several proteolytic enzymes are over-activated by high concentration of intracellular Ca^{2+}, roles of proteolytic enzymes associated with receptor-processing in NMDA-induced neuronal death are not fully understood. We investigated the effects of inhibition of proteolytic enzymes on NMDA-induced cell death. For analysis of cell viability, mitochondrial activity of primary rat cortical neurons was measured by XTT assay 24 hours after NMDA treatment with or without inhibitors of proteolytic enzymes. Inhibitors of matrix metalloproteinase, γ-secretase, protein convertase subtilisin / kexin type 9 and furin were examined whether they affect neuronal survival. Remarkable neuroprotective effect against NMDA-induced injury was observed in a dose-dependent manner by the furin inhibitor. Decreases in the amounts of NMDA receptor subunits and TrkB as well as intracellular signaling molecules Akt and glycogen synthase kinase 3 beta induced by NMDA treatment were suppressed by inhibitor of furin. Furthermore, the activation of calpain was significantly inhibited by the furin inhibitor. Thus, the activation of calpain would be involved in downstream of furin activation. These results indicated that the furin may be a therapeutic target for ischemic brain injuries.

Disclosures: **M. Yamada:** None. **H. Hayashi:** None. **N. Matsushima:** None. **M. Ohata:** None. **B. Yuan:** None. **N. Takagi:** None.

Poster

391. Molecular Mechanisms of Ischemia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 391.29/AA20

Topic: C.07. Ischemia

Support: NIH Grant GM107469

The Ralph S. French Charitable Foundation Trust

Title: *In vivo* studies of neuronal activity in the primary motor cortex after transient ischemic attack

Authors: *H. ZHOU[1], H. A. RIINA[2], J. ARD[1], A. ROSENBERG[1], G. YANG[1]
[1]Dept. of Anesthesiol., [2]Dept. of Neurosurg., New York Univ. Sch. of Medicine, NYU Langon, New York, NY

Abstract: A transient ischemic attack (TIA) is a brief, reversible episode of neurological dysfunction caused by focal ischemia without acute infarction. Evidence indicates that a transient interruption in cerebral blood flow may impair sensation, motor function and cognition. However, the effect of focal cerebral ischemia on brain activity at the cellular and subcellular level remains unclear. In this study, we mimicked TIA by producing a targeted ischemic attack in the primary motor cortex using photothrombosis. Using in vivo two-photon microscopy, we monitored the cortical activity before and after focal ischemia induction in real time by recording the Ca^{2+} activity of cortical pyramidal neurons that express genetically encoded Ca^{2+} sensor (GCaMP6s). Under normal conditions, pyramidal neurons in the motor cortex show increased somatic and dendritic Ca^{2+} activity when the animals are running on a treadmill. Following targeted focal ischemia in the superficial layer of the motor cortex, we observed a marked reduction of neuronal activity in both layer 2/3 and layer 5 pyramidal cells. This reduction of neuronal activity persists for at least 24 hours after the focal ischemia has been resolved. These results suggest that a transient interruption of cerebral blood flow may have long-lasting effect on neuron activity in local cortical circuitry. Currently, we are screening compounds that have the potential to mitigate the deleterious effects of focal ischemia on neuronal activity and motor function.

Disclosures: **H. Zhou:** None. **H.A. Riina:** None. **J. Ard:** None. **A. Rosenberg:** None. **G. Yang:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.01/AA21

Topic: C.09. Brain Injury and Trauma

Support: NIH R03NS095038 (S. Villapol)

NIH R01NS067417 (M.P. Burns)

Title: Targeting the liver acute phase response following traumatic brain injury reduces brain inflammation

Authors: E. WICKER[1], L. BENTON[2], S. ALAIYED[1], W. MUALEM[3], M. P. BURNS[3], *S. VILLAPOL[4]
[1]Pharmacol. and Physiol., [2]Dept. of Biochem. and Mol. Biol., [3]Dept. of Neurosci., Georgetown Univ., Washington, DC; [4]Departament of Neurosci., Georgetown Univ. Med. Ctr., Washington, DC

Abstract: Traumatic brain injury (TBI) triggers a loss of tissue followed by a strong and acute inflammatory response. Serum amyloid A1 (SAA1) is an acute phase protein that is mainly produced in the liver in response to inflammatory events such as TBI. SAA1 is released into the bloodstream, establishing a channel of communication between the liver and extra-hepatic organs. To date, no *in vivo* studies have investigated the role of SAA1 release in the central inflammatory response after TBI. We performed a mild and moderate/severe controlled cortical impact (CCI) injury in male C57BL/6J mice. One week before CCI, we injected antisense oligonucleotides (ASOs) (25 mg/kg/week by i.p.) to suppress mRNA SAA1 production. We wanted to explore the potential application of ASOs by modifying the inflammatory response. Western blot and ELISA analysis were used to quantify the expression of SAA1 in the serum and liver at several time points after CCI; immunohistological analysis was performed to identify macrophages/microglial and inflammatory cells binding SAA1 in the brain, *in situ* hybridization to detect mRNA SAA1 expression in the brain, and motor behavioral test were performed. Our results revealed that the highest grade of TBI severity was correlated with high levels of SAA1 in plasma at 1 dpi, and with a larger lesion volume and inflammation. We also found that SAA1 colocalizes with activated macrophages/microglial cells in the injured cortex and hippocampus; blocking SAA1 production reduced microglia activation and macrophage infiltration into the brain. Finally, we demonstrated that ASOs reduced SAA1 levels by 60-80% in plasma at 1 and 3 days after brain injury. ASO-treated mice displayed better motor skills on the rotarod 1 day after injury. Collectively, our findings demonstrate that SAA1 levels in plasma after brain damage depend on damage severity, acting as a potential biomarker. Also, we found that SAA1 binds to macrophages/microglial cells in the injured brain, and that ASO treatment reduces the

inflammatory response in the damaged brain. Altogether, this suggests that blocking SAA1 production could provide an advantageous environment for repair and functional recovery after brain trauma.

Disclosures: E. Wicker: None. **L. Benton:** None. **S. Alaiyed:** None. **W. Mualem:** None. **M.P. Burns:** None. **S. Villapol:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.02/AA22

Topic: C.09. Brain Injury and Trauma

Title: Effects of Apyrase in a rat model of cerebral contusion injury

Authors: *Y. FURUKAWA, M. KOBAYASHI, T. KUMAGAWA, K. SHIJO, N. MORO, M. FUKUSHIMA, T. MAEDA, A. YOSHINO
Neurosurg., Nihon Univ. Sch. of Med., Tokyo, Japan

Abstract: We have shown that gliotransmittion is activated immediately after traumatic brain injury. Gliotransmitter ATP is released into the extracellular space from injury stimulated astrocyte that will activate nearby microglia to initiate inflammatory response. In the previous study, we have shown that blockade of major receptor in gliotransmittion will suppress the activation of microglia and reduce the release of several cytokines in the extracellular space. In the present study we used Apyrase, an enzyme that catalyses the hydrolysis of ATP, to shut down the gliotransmittion and observed if it can suppress inflammatory response. Cerebral contusion injury was made in a rat and osmotic pump was implanted to deliver Apyrase or saline directly into the injured tissue. Rats were sacrificed at day 1, 3 and 7 following injury and analyzed. On day 1 and 3 after injury, increased level of activated microglia was observed by both histological staining and western blotting of Galectin-3. In the Apyrase injected group the activation was suppressed compared to the saline group in tissue near the contusion site. Few cytokine levels were suppressed by Apyrase injection in a mRNA level. Suppression of gliotransmittion by Apyrase have an effect to reduce inflammatory response after experimental cerebral contusion injury.

Disclosures: Y. Furukawa: None. **M. Kobayashi:** None. **T. Kumagawa:** None. **K. Shijo:** None. **N. Moro:** None. **M. Fukushima:** None. **T. Maeda:** None. **A. Yoshino:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.03/AA23

Topic: C.09. Brain Injury and Trauma

Title: Early reduction of large layer V corticospinal motor neurons precedes the inflammatory response and upregulation of phosphorylated-tau following repetitive mild TBI

Authors: *M. ALKASLASI, N. CHO, N. DHILLON, P. HARO-LOPEZ, O. SHELEST, G. BARMPARAS, E. LEY, G. M. THOMSEN
Cedars-Sinai Med. Ctr., West Hollywood, CA

Abstract: Objectives: Chronic traumatic encephalopathy (CTE) is a neurodegenerative disease linked to repeated head injury. The absence of stringent diagnostic tools together with variable disease progression limit our understanding of the disease. There are currently few experimental models that attempt to recapitulate the progressive brain atrophy and tau pathology that characterize the human condition. Using a recently described novel rat model of CTE, we examined temporal changes in layer V cortical neurons, the inflammatory response, and hyper-phosphorylated tau (P-tau) following repeat mild TBI.
Methods: Mild, bilateral, closed-skull TBI was administered once per week for 5 weeks, to rats at post-natal day (p) 60 once weekly for either 2 or 5 weeks. Sham and TBI rats were euthanized at acute (p90), short (p144) or long (p235) time points. TBI rats were classified as having “mild” or “severe” injuries based on rotorod performance following the last injury. Brain tissue was collected for stereological analysis to quantify layer V corticospinal motor neurons (CTIP2+ cells, large >300um), inflammatory cells (Iba1), and P-tau (AT8) over time.
Results: TBI rats exhibiting only mild rotorod deficits did not demonstrate a significant loss of large layer V corticospinal motor neurons at any time point following injury. Conversely, those with severe rotorod deficits exhibited a significant loss of large corticospinal motor neurons at the acute time point that persisted over time. Although there is an evident reduction of large corticospinal motor neurons in layer V, the total number of CTIP2+ cells in this region remained constant between sham and TBI groups at both the acute and long time points and between both mild and severe symptom groups. This would suggest that motor neurons are not dying but are perhaps unhealthy and shrinking due to injury. This apparent shrinkage of motor neurons preceded both the microglial inflammatory response and the upregulation of P-tau that was evident in both severity groups at the short and long time points, but not at the acute time point.
Conclusions: Following repeat mild TBI, rats that display severe motor deficits and progressive CTE-like brain pathology exhibit a significant reduction in large layer V corticospinal motor neurons that precedes the microglia response and upregulation of P-tau. This suggests that early targeting of neuronal cell damage could be beneficial to altering the progression of CTE caused

by repetitive head injury. By continuing to elucidate the underlying mechanisms of this disease, appropriate treatment strategies can be developed and applied.

Disclosures: **M. Alkaslasi:** None. **N. Cho:** None. **N. Dhillon:** None. **P. Haro-Lopez:** None. **O. Shelest:** None. **G. Barmparas:** None. **E. Ley:** None. **G.M. Thomsen:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.04/AA24

Topic: C.09. Brain Injury and Trauma

Title: Investigating the inflammatory response to midline and lateral fluid percussion injury

Authors: *O. KOKIKO-COCHRAN[1], K. WITCHER[1], D. EIFERMAN[2], J. GODBOUT[1]
[1]Neurosci., [2]Critical Care, Trauma, & Burn, Ohio State Univ., Columbus, OH

Abstract: Animal modeling studies have become essential in identifying molecular mechanisms driving post-traumatic brain injury (TBI) pathology as well as potential therapeutic targets to improve recovery. Fluid percussion-induced TBI is one of the most popular experimental models available. Typically, fluid percussion injury (FPI) is applied at midline or parasagittal to midline in rodents. Midline FPI is broadly defined as a diffuse model of TBI in which both hemispheres are equally injured. Lateral FPI is a mixed model of TBI in which the ipsilateral hemisphere receives a focal injury resulting in diffuse damage to subcortical structures. The present study sought to investigate the acute and sub-acute inflammatory response to midline and lateral fluid percussion injury (FPI), with specific emphasis on leukocyte recruitment and cytokine expression within the brain at acute and sub-acute time points. We hypothesized that the acute inflammatory response to TBI would be enhanced following lateral FPI compared to midline FPI resulting in hippocampal-dependent functional decline at sub-acute time points. To test this hypothesis, lateral or midline FPI or sham injury was administered to equal numbers of male and female C57BL6 mice at two months of age. By four hours post-injury, flow cytometric analysis revealed a TBI-induced increase in circulating neutrophils following midline and lateral FPI. Unexpectedly, sham injured mice receiving a midline or lateral craniectomy displayed a higher percentage of patrolling monocytes compared to TBI mice. Furthermore, both sham and TBI mice in the lateral FPI group displayed a higher percentage of circulating inflammatory monocytes. Brain neutrophils and inflammatory monocytes increased following midline and lateral FPI and occurred in conjunction with an increase in inflammatory cytokines IL1-β, TNF-α, and CCL2 at 4 hours post-injury. By 72 hours post-injury, the percentage of circulating neutrophils and monocytes declined in midline and lateral TBI mice. Similarly, brain leukocytes and inflammatory cytokine expression decreased by 72 hours post-injury, although CCL2 expression remained increased in the ipsilateral hemisphere following lateral FPI. Taken

together, these data indicate that the acute inflammatory response to midline and lateral FPI is quite similar with regard to leukocyte recruitment and cytokine expression soon after injury. Further studies are underway to determine differences in behavior and leukocyte recruitment at sub-acute time points.

Disclosures: O. Kokiko-Cochran: None. **K. Witcher:** None. **D. Eiferman:** None. **J. Godbout:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.05/AA25

Topic: C.09. Brain Injury and Trauma

Support: Merit Review Award #B1097-I

Title: Chronic inflammation and neurodegeneration after traumatic brain injury in swine

Authors: *M. R. GROVOLA[1,2], D. P. BROWN[1,2], K. D. BROWNE[1,2], C. E. KEATING[1,2], K. L. WOFFORD[2,3], J. A. WOLF[1,2], D. CULLEN[1,2], J. E. DUDA[2]
[1]Ctr. for Brain Injury and Repair, Dept of Neurosurg., Univ. of Pennsylvania, Philadelphia, PA; [2]Ctr. for Neurotrauma, Neurodegeneration, and Restoration, Corporal Michael J. Crescenz VA Med. Ctr., Philadelphia, PA; [3]Sch. of Biomed. Engin., Drexel Univ., Philadelphia, PA

Abstract: Every year in the US, 2.4 million people experience traumatic brain injury (TBI). Post-mortem human pathological studies have shown chronic inflammation and ongoing white matter degeneration after a single TBI. Moreover, a single moderate-to-severe TBI has been identified as a risk factor for Parkinson's disease (PD), Alzheimer's disease (AD), and other neurodegenerative dementias. The current study sought to examine chronic neuropathological changes after a single mild TBI in swine. Using a closed-head, rotation-acceleration injury model, swine were injured and survived for various time points out to 1 year post-injury. Pathological examinations included the entire brain, but focused on areas known to be at risk in TBI, such as the corpus callosum, periventricular white matter, and hippocampus. Sections of porcine tissue were stained for the amyloid precursor protein (APP), a marker for axonal pathology, GFAP (astrocytes), Iba-1 (microglia/macrophages), and Syn303 (α-Synuclein). Initial semi-quantitative assessments found subtle pathology in discrete anatomical regions. At one month post-injury, specimens had significant reactive microglia and astrocytes in the hippocampus, subcortical white matter, and periventricular white matter. Additionally, one-month injured specimens had occasional APP+ dystrophic neurites in the periventricular white matter and corpus callosum. α-Synuclein (α-Syn) also appeared to be upregulated in several regions of the thalamus, which was reflected by increased somatodendritic Syn303 staining in

some cells and the emergence of α-Syn over-expression in a subset of white matter tracts. In the one-year cohort, time-matched shams exhibited reactive microglia in the hippocampus and periventricular white matter, but not in subcortical white matter; astrocytes exhibited normal phenotypes in these regions. There was no noticeable APP or α-Syn pathology in one-year sham specimens. In contrast, one year post-injury specimens had significant reactive microglia and astrocytes in the hippocampus, subcortical white matter, and periventricular white matter; however the amount of reactive microglia in the white matter was noticeably less than at 1 month post-injury. There was no noticeable APP pathology at one year post-injury, although thalamic α-Syn over-expression appeared to increase in presence and intensity. These results suggest that persistent inflammation and alterations in α-Syn expression may occur after a single TBI. These findings provide insight to neurodegenerative disease as a sequela to TBI, particularly as neuroinflammation is increasingly linked to the development of AD and PD.

Disclosures: M.R. Grovola: None. **D.P. Brown:** None. **K.D. Browne:** None. **C.E. Keating:** None. **K.L. Wofford:** None. **J.A. Wolf:** None. **D. Cullen:** None. **J.E. Duda:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.06/AA26

Topic: C.09. Brain Injury and Trauma

Support: Swedish Armed Forces R&D (AF.9221006)

Title: Neurofilament light concentrations in serum following experimental rotational traumatic brain injury in rats

Authors: *M. G. RISLING[1], M. ANGÉRIA[2], H. ZETTERBERG[3], K. BLENNOW[3], J. DAVIDSSON[4]
[1]Karolinska Inst., Stockholm S-171 77, Sweden; [2]Neurosci., Karolinska Institutet, Stockholm, Sweden; [3]Clin. Neurochemistry Lab., Sahlgrenska Univ. Hosp., Mölndal, Sweden; [4]Dept. of Mechanics and Maritime Sci., Chalmers Univ. of Technol., Göteborg, Sweden

Abstract: The protein neurofilament light (NFL) is predominantly expressed in the long myelinated subcortical axons. Measurement of NFL in serum of athletes after head trauma have been shown to correlate with the severity of brain injury. However, experimental models that allow for studies of NFL in serum in combination with studies of structural brain injuries are limited. Therefore, the aim of this study was to measure NFL in serum from rats following experimental sagittal plane rotation trauma and to compare these measurements to data on accumulation of Amyloid Precursor Protein (APP) in the white matter following similar type and severity trauma. Spray Dawley rats (n=65) were exposed to rapid sagittal plane rotational

acceleration head trauma (maximum rotational acceleration 1.40 ± 0.32 (mean ± SD, n=29). Serum samples were collected just prior to termination at 2.5, 24, 72, 120, 168 and 672 hours post trauma. The NFL serum concentration was studied using NFL immunoassay on the single molecule array (Simoa) platform. The trauma caused subdural bleedings in most animals. Staining brain tissue with APP antibodies and FD Neurosilver revealed wide spread axonal injuries in the corpus callosum, the border between the corpus callosum and cortex and in tracts in the brain stem. Only limited signs of contusion injuries were observed following trauma. Macrophage invasions, glial fibrillary acidic protein redistribution or hypertrophy, and blood brain barrier (BBB) changes were unusual. Traumatized animals exhibited increased NFL concentrations compared to sham exposed animals for all survival times; for survival times 24 h the increase was 8 times (exposed 666 pg/mL (range 327 to 1421 pg/mL) sham 83 pg/mL (range 40 to 114 pg/mL). The shorter and longer survival times resulted in lesser increase. At 2.5 hours and 28 days post trauma the NL serum concentration increase were approximately 2.5 times. The measured NFL leakage to the blood system appear to occur despite the absence of obvious BBB injuries. Serum NFL concentration appear to correlate with APP level in the white matter for all time points studied. In conclusion NFL serum concentrations in experimental trauma was more prominent than those reported following sports-related concussion and when using the same immunoassay technique. This study supports the hypothesis that mild rotational trauma is associated with acute axonal injury and serum NFL may offer diagnostic utility for such trauma. NFL serum level increased following trauma despite no apparent BBB injury.

Disclosures: **M.G. Risling:** None. **M. Angéria:** None. **H. Zetterberg:** None. **K. Blennow:** None. **J. Davidsson:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.07/AA27

Topic: C.09. Brain Injury and Trauma

Support: CDMRP G1701725

Title: Divergent glial response following CCI versus blast injury in the gyrencephalic ferret brain

Authors: *S. C. SCHWERIN[1], M. CHATTERJEE[1], E. B. HUTCHINSON[2], K. RADOMSKI[4], A. IMAM-FULANI[1], J. T. MCCABE[5], C. PIERPAOLI[3], S. L. JULIANO[1]
[1]Anatomy, Physiol. and Genet., Uniformed Services Univ., Bethesda, MD; [3]Quantitative Med. Imaging Section, NIBIB, [2]NIH, Bethesda, MD; [4]Ctr. for Neurosci. and Regenerative Med., Henry M. Jackson Fndn., Bethesda, MD; [5]Dept. of Anatomy, Physiol. & Genetics, Uniformed Services Univ., Bethesda, MD

Abstract: The ferret brain is gyrencephalic, has a high white to gray matter ratio, and the placement of the hippocampus is in a ventral location similar to humans. The pathological mechanisms occurring after TBI in the ferret may therefore be more similar to humans than previously reported findings in rodents. In this study we investigated markers of injury after two different types of TBI: controlled cortical impact (CCI) and blast. Adult male ferrets either received a mild CCI (velocity = 5 m/s, depth = 2 mm, impactor diameter = 3mm, dwell time = 100 ms) in the somatosensory cortex, a single mild blast (15-17 psi) or a quick succession of 4 mild blasts (15-17 psi each, occurring within 30 minutes) and survived for various times after injury. We evaluated pathological markers of brain injury using immunohistochemistry (IBA-1: microglia; GFAP: astrocytes; CP13: phosphorylated Tau) and behavior (open field, novel object recognition, Tmaze, gait). RESULTS. A focal increase in astrocytes occurred in the region of CCI injury - both in the cortex and immediate underlying white matter. In the animals receiving a blast, however, there was a global upregulation of astrocytes just beneath the pia, in the sulcal depths, and around blood vessels in the neocortex, as well as a displaying a strong pattern of astrogliosis in the white matter. There was little to no change in microglial morphology or density in the brain after blast. In the CCI animals, however, microglia were found in clusters in the white matter extending several millimeters away from the center of the injury. Markers for phosphorylated tau were increased within the brain after blast injury but were relatively unchanged after CCI. Motor behavior was subtly affected after mild CCI, but not at all after 4X blast. Cognitive skills such as memory began to decline at 4 weeks after injury in both CCI and blast animals. In summary, two types of brain injury were studied in a gyrencephalic animal model which expressed different spatial/temporal pathological responses across glial subtypes. The astrocyte response appeared similar to that observed in humans and supports the use of the ferret in studying traumatic brain injury.

Disclosures: S.C. Schwerin: None. **M. Chatterjee:** None. **E.B. Hutchinson:** None. **K. Radomski:** None. **A. Imam-Fulani:** None. **J.T. McCabe:** None. **C. Pierpaoli:** None. **S.L. Juliano:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.08/AA28

Topic: C.09. Brain Injury and Trauma

Support: Intramural Combat Casualty Care Research Program (CCCRP) Grant.

Title: Comprehensive dose-response study of progesterone in a preclinical model of penetrating ballistic-like brain injury

Authors: *C. M. GROEBER TRAVIS, S. L. OKADA-RISING, R. C. PEDERSEN, J. A. SUN, K. E. DEDOMINICIS, D. A. SHEAR
BTNN, Walter Reed Army Inst. of Res., Silver Spring, MD

Abstract: Progesterone has shown great promise in preclinical studies as a potent anti-edemic and neuroprotective therapy for traumatic brain injury (TBI). However, a recent Phase III clinical trial for progesterone as a treatment for TBI was recently stopped for futility. While there are a number of reasons why success achieved at the preclinical level with progesterone failed to translate to the clinical level, it may be partly due to the dosing regimen. In the current study, we tested 4 dose concentrations (1, 5, 10 and 20 mg/kg) for neurobehavioral outcomes in a rodent model of penetrating ballistic-like brain injury (PBBI). Unilateral frontal PBBI (10%) was induced in the right hemisphere of isoflurane anesthetized rats. Progesterone (22.5% 2-hydroxylpropyl-B-cyclodextrin) was administered as 10 minute intravenous infusions delivered at 30 min and 6 hours post-PBBI and then every 24 hours for 5 consecutive days. Results showed that 5 mg/kg was effective in reducing cognitive deficits in the Morris water maze (MWM) but was not effective in reducing motor impairment on the rotarod task. Conversely, 10 mg/kg improved motor outcome but did not ameliorate cognitive deficits. Additional experiments assessed the effects of progesterone (5 mg/kg) on reducing brain edema (wet/dry weights) at 24, 48, and 72 hours post-PBBI. Overall, progesterone failed to mitigate brain edema following PBBI. However, in brain regions distal from the lesion core, progesterone did appear to be improving recovery from edema at 48h post-injury. Additional post-injury time points are being evaluated and will be presented at the meeting. Collectively, the current results support the preclinical literature indicating that progesterone is effective in ameliorating behavioral decrements in a model of severe penetrating TBI but dosing regimens may need to be tailored more specifically to injury severity.

Disclosures: C.M. Groeber Travis: None. **S.L. Okada-Rising:** None. **R.C. Pedersen:** None. **J.A. Sun:** None. **K.E. DeDominicis:** None. **D.A. Shear:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.09/AA29

Topic: C.09. Brain Injury and Trauma

Title: Transcriptional alterations following traumatic brain injury in mOHSC model

Authors: *D. ZHOU[1], H. YAO[1], I. HARTLEY[1], J. WANG[1], W. WU[1], O. POULSEN[1], J. XUE[1], G. G. HADDAD[1,2,3]
[1]Pediatrics, UCSD, La Jolla, CA; [2]Neurosciences, Univ. of California San Diego, La Jolla, CA; [3]The Rady Children's Hosp., San Diego, CA

Abstract: Traumatic brain injury (TBI) may occur at any time and age. It is the leading cause of disability and death in children and adolescents in the US. The nature of the injury and its consequences vary and the course of recovery is very difficult to predict. Even though early diagnosis and ongoing therapeutic intervention may decrease the severity of symptoms, TBI, and subsequent secondary brain injuries (SBI), may still lead to long lasting impairments in one or more brain functions (such as cognitive function, physical abilities, communication and/or social/behavioral aspects). It may also trigger or predispose subjects to a variety of neuronal disorders. Therefore, a better understanding of the molecular mechanisms underlying TBI injury is essential for identifying potential targets for novel therapeutic strategies. To do so, we developed a mouse organotypic hippocampal slice culture injury model (mOHSC) in which a specified stretch for a specified duration was introduced to hippocampal slices grown on silastic membranes of the Bioflex plate to mimic the stretch impact in TBI. The level of injury was measured using PI staining of the dead cells to monitor the level of injury following stretch. Using RNA-seq, we investigated gene expression 2 hours after the introduction of the stretch. We detected a total of 265 genes with significant alteration in expression (77 were up- and 188 were down-regulated). These alterations demonstrated the presence of various pathological changes, including apoptosis, necrosis, brain edema, leakage of blood-brain barrier, severe astrocytosis, development of glial scar, leukocyte migration and tauopathy. Furthermore, these transcriptional changes also suggest potential predispositions for a variety of neuronal disorders following TBI injury, including neurodegeneration and Alzheimer disease. The current study demonstrated that mOSHC is a convenient and powerful model to study the mechanisms underlying immediate as well as consequential molecular responses to traumatic brain injury.

Disclosures: D. Zhou: None. **H. Yao:** None. **I. Hartley:** None. **J. Wang:** None. **W. Wu:** None. **O. Poulsen:** None. **J. Xue:** None. **G.G. Haddad:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.10/AA30

Topic: C.09. Brain Injury and Trauma

Support: College of Pharmacy seed grant

Title: Temporal changes in mast cell release following a mild traumatic brain injury in rats

Authors: *H. O. AWWAD[1], M. R. LERNER[2], M. BAIER[3]
[1]Pharmaceut. Sciences, Col. of Pharm., [2]Surgery, Univ. of Oklahoma Hlth. Sci. Ctr., Oklahoma City, OK; [3]Pharmaceut. Sciences, Col. of Pharm., Univ. of Oklahoma Hlth. Sci. Ctr., Oklahoma city, OK

Abstract: Following a TBI, shear force and stress to brain tissue releases a plethora of neurotoxic factors in addition to glutamate that contribute to TBI pathology and a worse prognosis, if left untreated. However, histamine release in cerebral ischemia alleviates neuronal apoptosis and improves neurological function and histamine receptors are implicated in neuronal disorders to modulate memory, pain and sleep. The specific objective of this study was to quantify the release of histamine and to correlate these changes with pain and vestibulomotor deficits following a mild TBI (mTBI) in Sprague-Dawley male rats compared to sham rats. Brains were extracted from rats euthanized on day 1 or day 8 post-TBI using the controlled cortical impact model. Levels of injury proteins and mast cells were quantified in the ipsilateral (left) and contralateral (right) sensory cortex (SC) using immunoblotting analysis and immunofluorescence microscopy. Rotarod performance, tactile allodynia and thermal hyperalgesia were assessed up to day 8 post-injury/sham. mTBI-induced vestibulomotor deficits were transient and rats showed complete recovery within 7 days of injury, whereas the same animals did not recover from mTBI-induced neurological deficits, tactile allodynia and thermal hyperalgesia during that same post-injury period. Reactive astrogliosis and degranulated mast cells were detected in mTBI rats at day 1 post-injury using immunohistochemistry. In conclusion, changes in histamine levels seem to correlate with the histopathology of reactive astrogliosis and neuroinflammation as well as increased pain sensitivity following a mild TBI.

Disclosures: H.O. Awwad: None. **M.R. Lerner:** None. **M. Baier:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.11/AA31

Topic: C.09. Brain Injury and Trauma

Support: NIH-NS40125

NIH-NS060672

VAI01RX001127

NIH 1-F32NS090748

The Pittsburgh Foundation

Title: Synaptic formation of alpha synuclein aggregates acutely following experimental traumatic brain injury

Authors: *C. DIXON, Y. LI, J. HENCHIR, X. MA, S. W. CARLSON
Neurosurg., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Traumatic brain injury (TBI) has been identified as a risk factor for the development of chronic neurodegenerative diseases, including Parkinson's disease. Pathological protein misfolding is associated with disrupted cellular function and can contribute to subsequent neuronal loss. Current hypotheses suggest that synaptic dysfunction precedes neuronal loss in the pathological progression of neurodegenerative disease. Alpha synuclein (α-synuclein) has been well described for its role in contributing to the pathogenesis of Parkinson's disease. Pathological forms of α-synuclein have been observed at chronic time points following TBI. Emerging evidence highlights a central role for α-synuclein in the formation of the soluble N-ethylmaleimide-sensitive factor attachment protein receptor (SNARE) complex responsible for neurotransmitter release. We recently reported that TBI results in an acute reduction in monomeric α-synuclein. We hypothesized that TBI results in acute formation of α-synuclein aggregates in synapses. To this end, male Sprague Dawley rats were subjected to sham or controlled cortical impact (CCI) injury (2.7 mm, 4 m/s, 150 msec) and sacrificed at 2, 7, or 14 days post-injury. High molecular weight α-synuclein aggregates, confirmed using two antibodies, were detected in hippocampal whole cell and synaptosomal-enriched lysates at all time points assessed. Immunohistochemical staining revealed aggregated α-synuclein immunoreactivity in pericontusional gray and white matter and the dentate gyrus of the hippocampus at 2 weeks following CCI injury. These findings provide novel evidence for acute α-synuclein aggregates in the injured synapse that may contribute to impaired synaptic and neurobehavioral function after TBI.

Disclosures: C. Dixon: None. **Y. Li:** None. **J. Henchir:** None. **X. Ma:** None. **S.W. Carlson:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.12/AA32

Topic: C.09. Brain Injury and Trauma

Support: Supported by Bill and Melinda Gates Foundation Cysticercosis Elimination in Peru grants 23981

NIH grant 5D43TW006581 (Infectious Diseases Training Program in Peru) (R.H.G.)

NIH training grant FIC/NIH D43 TW001140

Fondecyt 118-2015

INNOVATE Peru 2015

Title: Neuroinflammation and Neuronal damage in neurocysticercosis using animal model

Authors: *M. R. VERASTEGUI[1], R. P. CARMEN[1], R. H. GILMAN[3], D. G. DÁVILA[1], G. CASTILLO[1], J. D. MORALES[1], N. CHILE[1], E. G. BERNAL[1], B. J. CONDORI[1], A. D. DELGADO[1], L. E. BAQUEDANO[1], C. CYSTICERCOSIS-WORKING-GROUP[2]
[1]Infectious Dis. Lab. Research-LID, [2]Univ. Peruana Cayetano Heredia, Lima, Peru; [3]Bloomberg Sch. of Hyg. and Publ. Hlth., Johns Hopkins Univ., Baltimore, MD

Abstract: Human neurocysticercosis (NCC) is a parasitic disease caused by larvae of the cestode *Taenia solium* (cysticercus), when invade central nervous system (CNS). Human NCC is one of the most significant parasitic diseases of the CNS. It is an important contributor to neurological morbidity in developing countries, and is the major cause of acquired epilepsy in the developing world. We developed a NCC animal model using rats, which is useful tool to investigate NCC pathology. The following study describes the neuronal damage in rats infected with activated oncospheres of *T. solium* via two routes of infections: direct intracranial injection and oral feeding. At 4-5 months after infection where the larvae is mature, infected brains present extensive axonal spheroids immunoreactive to neurofilament and amyloid precursor protein. We did not observe difference in the frequency nor the average number of axonal spheroids in both routes of infection. This pathology is observed in conjunction with perivascular inflammatory cuffing (PIC), edema and gliosis in the tissue adjacent to the parasite as well as extending beyond this area into otherwise appearing normal brain tissue. Gliosis (positive to GFAP and Iba-1) were similar in both routes but PIC and inflammatory cells (heterophils and plasmocytes) were significantly higher in the intracranial route. Taken together, our data provides the first evidence that neuronal lesion may contribute to the pathogenesis in experimental NCC. Additionally, our findings may represent an interesting avenue for future study in this parasitic brain disease for their potential as a common crossroad between neuroinfectious diseases such as NCC and neurodegenerative diseases, which classically exhibit neuronal damage.

Disclosures: M.R. Verastegui: None. **R.P. Carmen:** None. **R.H. Gilman:** None. **D.G. Dávila:** None. **G. Castillo:** None. **J.D. Morales:** None. **N. Chile:** None. **E.G. Bernal:** None. **B.J. Condori:** None. **A.D. Delgado:** None. **L.E. Baquedano:** None. **C. Cysticercosis-Working-Group:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.13/AA33

Topic: C.09. Brain Injury and Trauma

Support: CIHR

NSERC

OICR

Title: Sex modulates the impact of inflammation on brain development after cranial irradiation

Authors: *E. DE GUZMAN[1,2], M. AHMED[1], S. PERRIER[1], B. NIEMAN[1,2,3]
[1]Mouse Imaging Ctr., Hosp. For Sick Children, Toronto, ON, Canada; [2]Dept. of Med. Biophysics, Univ. of Toronto, Toronto, ON, Canada; [3]Ontario Inst. for Cancer Res., Toronto, ON, Canada

Abstract: Introduction: Cranial radiation therapy (CRT) is one of the main contributing factors to long term neurocognitive deficits in survivors of pediatric cancers. The risk for and severity of these deficits are greater in female patients. CRT also induces neuroanatomical changes detectable as volume deficits with magnetic resonance imaging (MRI). These are likewise more pronounced in females, both in humans [1] and, to a lesser extent, in mice [2]. While the direct cellular cause of these deficits remains to be elucidated, it has been hypothesized that changes to the cellular environment post-irradiation alter neural progenitor cell fate, and may inhibit recovery [3]. Irradiation induces a chronic inflammatory response that may be responsible [4]. With strong evidence suggesting sex is an important factor in immune response [5], the goal of this study was to determine how irradiation induced changes in neuroanatomical development depend on inflammatory response, and whether or not that response is dependent on sex.
Methods: C-C chemokine ligand 2 (CCL2; previously called monocyte chemoattractant protein, MCP-1) has been shown to play a critical role in neuroinflammation [6]. Knockout of its receptor, CCR2, recovers neurogenesis in the irradiated adult mouse brain and is associated with recovery of hippocampal dependent behaviour [7]. As a result, we chose to irradiate CCL2-deficient and CCL2-wild-type mice with a whole brain dose of either 0 or 7 Gy during infancy. In vivo MRI was used to track brain development for 3 months following irradiation, and deformation-based morphometry was used to identify volume differences across development.
Results: Irradiation of wild type mice results in a deficit in neuroanatomical growth that is subtly more pronounced in females. Male CCL2-deficient mice are significantly protected from this damage, while female CCL2-deficient mice are not. The development of unirradiated CCL2-deficient mice is also different from wild type mice, confounding the response to irradiation. Our conclusions are two-fold: 1) immune response plays an important role in the development of irradiation induced neuroanatomical deficits, and 2) sex modulates the role of the immune response to radiation and is critical to consider in development of treatment strategies.
[1] BJ Nagel et al. (2004) AJNR Am J Neuroradiol. **[2]** AE de Guzman et al. (2015) Radiate Res. **[3]** ML Monje et al. (2002) Nat Med. **[4]** CS Wong et al. (2004) Mol Interv. **[5]** S Klein and K Flanagan (2016) Nat Rev Immunol **[6]** S Ge et al. (2009) J Mol Neurosci **[7]** K Belarbi et al. (2013) Cancer Res.

Disclosures: E. De Guzman: None. **M. Ahmed:** None. **S. Perrier:** None. **B. Nieman:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.14/AA34

Topic: C.09. Brain Injury and Trauma

Support: NIH Grant HD001097

Cincinnati Children's Hospital Division of Pediatric Rehabilitation Medicine

Title: Optic tract degeneration and neuroinflammation in a closed head adolescent mouse model of traumatic brain injury

Authors: *N. K. EVANSON, F. GUILHAUME-CORREA
Div. of Pediatric Rehabil. Med., Cincinnati Children's Hosp. Med. Ctr., Cincinnati, OH

Abstract: Objective: To determine the nature of injury in adolescent mice after experimental closed head weight drop traumatic brain injury (TBI).
Methods: 6 week old male C57BL/6J mice were subjected to weight drop using a 400g rod and drop height of 1.5 cm under isoflurane anesthesia, or to anesthesia only. Separate cohorts of mice (n=8 per experimental group) were perfused either 24 hours or 7 days after injury, and brain was examined using histologic staining (NeuroSilver or FluoroJade degeneration stains, or Nissl Stain), or by immunohistochemistry to markers for neurons (NeuN), astrocytes (Glial Fibrillary Acidic Protein), microglia/macrophages (Iba-1), or a marker of axonal damage (amyloid precursor protein, APP). Other animals (n=16 per group) were subjected to experimental TBI, then underwent behavioral analysis including visual cliff, open field, and novel object recognition tests. The effect of coronal load on the skull was analyzed by MicroCT imaging. Planned experiments will test the effect of chronic variable stress on behavioral outcomes after experimental TBI.
Results: Gross pathology and Nissl stain did not reveal evidence of gross neurologic damage, and there was no widespread neuronal loss by NeuN staining. Fluoro Jade and NeuroSilver stains revealed evidence of axonal degeneration that was most evident in the bilateral optic tracts, the lateral geniculate nucleus of the thalamus, and superior colliculus. This was accompanied by astrocytic gliosis in these areas, and neuroinflammation as revealed by morphologic activation of microglia. MicroCT analysis of the mouse skull under dorsal-ventral load revealed partial collapse of the optic canal. Behavioral differences between TBI and sham animals were relatively subtle, and did not appear to demonstrate severe visual deficits.
Conclusions: Experimental closed head TBI in adolescent mice leads to reproducible damage to the optic tracts and proximal target regions, which is associated with astrocytosis and neuroinflammation. This damage appears to be caused by an optic nerve contusion due to collapse of the optic canal during TBI.

Disclosures: **N.K. Evanson:** None. **F. Guilhaume-Correa:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.15/AA35

Topic: C.09. Brain Injury and Trauma

Support: UTHealth Brain Initiative (Soto)

Alzheimer's Association NIRG-394284

Title: The effect of repetitive mild traumatic brain injury on tau pathology

Authors: *G. EDWARDS, III, N. MENDEZ DINAMARCA, C. SOTO, I. MORENO-GONZALEZ
Neurol., Univ. of Texas Hlth. Sci. Center-Houston, Houston, TX

Abstract: Mild traumatic brain injury is the most common form of concussion and is prevalent among contact sports athletes and military personnel. Repetitive mild traumatic brain injury (rmTBI) can trigger the activation of several molecular cascades that lead to neuropathological consequences in the brain. Moreover, rmTBI has been correlated to neurodegenerative diseases such as chronic traumatic encephalopathy (CTE). CTE is a slow, progressive tauopathy that exhibits neurofibrillary tangles (NFTs) accompanied by psychiatric and cognitive manifestations. NFTs are intracellular aggregates formed from the misfolding and aggregation of tau protein. A vast majority of individuals following a TBI event demonstrate early tau accumulation, independent of age, suggesting a potential pathological link between TBI and tauopathies. The role of tau aggregation in brain and in subsequent clinical symptoms following rmTBI remain to be elucidated. Moreover, effective diagnostic methods for TBI and tau pathology are lacking. The goal of the current study is to assess the generation of tau aggregates and brain pathology following rmTBI induction in mice. Thus, transgenic (Tg) tau mice were subjected to rmTBI events, which mimic multiple sub-concussive impacts in athletes, over various time-points. Tg tau mice subjected to rmTBI demonstrated neurobehavioral changes as well as early misfolded tau deposition and elevated tau levels in brain. These results indicate that rmTBI could be a trigger for early aggregated tau formation; in addition, misfolded tau can play a role in the psychiatric and cognitive behaviors affiliated to TBI and disease. Overall, this work will shed light to understanding the effect of rmTBI in relation to tau pathology and behavioral impairment and will potentially provide a potent diagnostic tool to detect tauopathy onset and monitor TBI-induced brain damage in the future.

Disclosures: **G. Edwards:** None. **N. Mendez Dinamarca:** None. **C. Soto:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Amprion. **I. Moreno-Gonzalez:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.16/AA36

Topic: C.09. Brain Injury and Trauma

Support: HU0001 15 2 0024

Title: Pial arteriolar hyper-responsiveness after blast was related with changes in endothelin-1 and endothelin receptors expression

Authors: *S. H.-E.-R. MULLAH[1], R. ABUTARBOUSH[1], Y. CHEN[1], M. LASHOFF-SULLIVAN[1], F. ARNAUD[1,2], U. KAWOOS[1], R. MCCARRON[1,2], S. T. AHLERS[1]
[1]Naval Med. Res. Ctr., Silver Spring, MD; [2]Uniformed Services Univ. of Hlth. Sci., Bethesda, MD

Abstract: Background: Traumatic brain injury (TBI) resulting from exposure to blast overpressure (BOP) component of improvised explosive devices (IEDs) is a prevalent injury in recent conflicts. Changes in cerebral vasculature, such as delayed presentation of cerebral vasospasm and blood-brain barrier dysfunction, have been reported following blast-induced TBI that present challenges to neurosurgical care and to the recovery of the patient. We previously reported functional changes in cerebrovascular responses within 24 hours after different BOP intensities using a cranial window technique in rodents. In this study, we examined vascular responsiveness over a longer timeframe after the blast exposure than our previous study and further explored potential underlying role of endothelin-related pathways after BOP exposure as a possible mechanism underlying BOP-induced cerebrovascular changes. **Methods:** Anesthetized male Long Evans rats were exposed to single blasts of either 0 (sham control), 37, 75 or 140 kPa while located inside a shock tube apparatus and then allowed to recover fully. At 2h, 1, 3, 14 or 28 days, rats were re-anesthetized, ventilated and a cranial window was made in the parietal bone to expose the surface of the cerebral cortex. Pial arteriolar diameters were measured in real time using intravital microscopy before and after hypercapnia (7.5% CO_2), and topical application of barium chloride (5% $BaCl_2$,) and serotonin (5-hydroxytryptamine, 5HT). The expression of endothelin-1 (ET-1), ETA and ETB receptors was examined in the frontal cortex at 1 and 3 days post-blast. **Results:** After blast, increases in vasoconstriction by $BaCl_2$ ($P < 0.01$), and vasodilatation by CO_2 were seen at all intensities of BOP. While control animals showed vasoconstriction by 5-HT, BOP-exposed animals showed vasodilation. The changes in pial arteriolar reactivity varied with blast intensity and time after blast. The increase in the

arteriolar response in BOP-exposed rats in the 37 kPa group appeared to normalize to baseline levels by 28 days. In the 140 kPa group increased responsiveness persisted until at least 28 days post-BOP. Preliminary RT-PCR data showed an increase in mRNA expression for ET-1 one day following low intensity blast. Western blot analysis indicated significantly lower levels of ETA receptor in the 140 kPa group and significantly lower levels of ETB receptor in 75 and 140 kPa groups at 3 days post-blast. **Conclusion:** Cerebral pial arteriolar hyper-responsiveness may correlate with BOP intensity. Changes in expression of ET-1, ETA and ETB receptors indicate a possible association with BOP-induced changes to cerebrovascular function.

Disclosures: S.H. Mullah: None. **R. Abutarboush:** None. **Y. Chen:** None. **M. Lashoff-Sullivan:** None. **F. Arnaud:** None. **U. Kawoos:** None. **R. McCarron:** None. **S.T. Ahlers:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.17/BB1

Topic: C.09. Brain Injury and Trauma

Support: MOST-105-2314-B-532 -008 -

Title: Nogo receptor-1 regulates balance, cognition and emotion after mild traumatic brain injury in mice

Authors: *J.-H. LAI, ESQ[1], K.-Y. CHEN[2], J. C.-C. WU[3], C.-Z. CHI-ZONG HUANG[3], Y.-H. CHEN[3], L. OLSON[4], Y.-H. CHIANG[3]
[1]Ph.D. Program for Neural Regenerative Med., [2]Grad. Inst. of Neural Regenerative Medicine, Col. of Med. Sci. and Technol., [3]Dept. of Surgery, Col. of Med., Taipei Med. Univ., Taipei, Taiwan; [4]Karolinska Inst., Stockholm, Sweden

Abstract: Background: Traumatic brain injury (TBI) is a major public health issue of the lesioned central nervous system. Among all the TBI, Mild TBI (mTB) has 75~90% of the TBI and can cause the various physical, cognitive, emotional, and psychological-related symptoms. Nogo receptor 1 (NgR1) is a glycosylphosphatidylinositol (GPI)-anchored protein on the cell surface and involved in the central nervous system (CNS) axonal plasticity, development, and recovery from injury. But the role of the NgR1 in behavior after mTBI is still unknown. The study focusses on investigating the role of NgR1 in the balance, cognition and emotion after mTBI in mice
Methods: NgR1-Tg mice were obtained from the Olson Laboratory in Karolinska Institute Stockholm, Sweden. NgR1-Tg mice were generated by CamKII-tTA mating with pTRE-NgR1 to obtain NgR1 overexpressing mice. Doxycycline (Sigma-Aldrich) dissolved in drinking water

(100 μg/mL) was giving to turn off transgene expression to mice carrying both the CamKII-tTA and the pTRE-NgR transgenes. Animal study were approved by the Institutional Animal Care and Use Committee (IACUC) of the Taipei Medical University and National Defense Medical Center. mTBI was induced by weight drop impact. Mice were anesthetized with isoflurane and then brain was placed soft platform and impacted with a rod-shaped 25-g metal dropped trough the guide tube (diameter 13 mm × length 80 cm).
Results: To determine the role NgR1 on behavior after mTBI, we tested NgR1-Tg mice by the four groups after non mTBI and mTBI with doxycycline or not. We found the NgR1-Tg mice with doxycycline were better than the NgR1-Tg mice without doxycycline after mTBI on balance, learning memory, cognition and anxiety. The behaviors after mTBI were recovery on the second week.
Conclusions: These results were shown that NgR1-Tg mice have the long-term memory, cognitive and emotional problems on non-mTBI mice but not in balance. The NgR1-Tg mice after mTBI were worse than these mice with doxycycline on balance, the long-term memory, cognitive and emotional. NgR1 could play an important role in physical, cognitive, and psychological-related behavior. Key words: mTBI, NgR1, behavior

Disclosures: J. Lai: None. **K. Chen:** None. **J.C. Wu:** None. **C. Chi-Zong Huang:** None. **Y. Chen:** None. **L. Olson:** None. **Y. Chiang:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.18/BB2

Topic: C.09. Brain Injury and Trauma

Support: DOD W81XWH-15-1-0303

Title: Blast-induced traumatic brain injury displays a unique pattern of spatial neuropathology

Authors: *N. CHANDRA[1], R. R. KAKULAVARAPU, 07102-1982[2], D. Y. YOUNGER, 07102-1982[2], A. A. ARAVIND, 07102-1982[2], B. P. PFISTER, 07102-1982[2], M. KURIAKOSE[2], M. SKOTAK[2]
[1]Biomed. Engin., New Jersey Inst. of Technol., Morris Plains, NJ; [2]Biomed. Engin., New Jersey Inst. of Technol., Newar, NJ

Abstract: Blast-induced Traumatic brain injury (bTBI) is a leading cause of morbidity and mortality in soldiers in the combat and in training sites and is becoming an important public health issue. The bTBI simultaneously causes primary (shockwave), secondary (penetrating), and tertiary (blunt-accelerative forces) injuries causing different loading modalities and pathological outcomes compared to pure blunt injury. The brain injury patterns resulting from primary

shockwave propagation across different brain structures has not been carefully investigated. We hypothesize that bTBI has a unique histopatholgy and pathophysiology different from blunt TBI. In bTBI all the regions of brain from prefrontal cortex to hippocampus to cerebellum to brain stem are all simultaneously biomechanically loaded as the shock wave propagates unimpeded across the brain. This is different from the blunt TBI, where the injuries are mostly confined to the site of injury and nearby areas, the extent of which is dictated by the severity of blunt loading. Since oxidative stress has been implicated as a major pathogenic factor in TBI, here we examined the brain spatial resolution and cellular distribution of changes in superoxide producing enzyme NADPH oxidase1 (NOX1) as well as neurodegeneration paradigm during the early phase (4h) of moderate blast TBI (180 Kpa) and compared these data with that of moderate blunt TBI (FPI). Immunofluorescence analysis displayed that NOX1 was upregulated in different brain regions and showed a differential vulnerability with the highest increase found in hippocampus and thalamus followed by frontal cortex and cerebellum. Among different cells, neurons exhibit highest amount of basal NOX1 and its increase was higher in hippocampal and cerebellar regions compared to astrocytes and microglia. Interestingly, blunt injury induced in the cortical region did not display any change in NOX1 expression in cerebellum suggesting that the spread of the injury in blunt TBI is localized. Blast did not display neurodegeneration in any region, while blunt TBI showed focal neurodegeneration in the cortical and hippocampus areas only in ipsilateral side. These studies indicate that the injury pattern and pathological changes in blast TBI are unique and spread across the entire brain compared to blunt injuries.

Disclosures: **N. Chandra:** None. **R.R. Kakulavarapu:** None. **D.Y. Younger:** None. **A.A. Aravind:** None. **B.P. Pfister:** None. **M. Kuriakose:** None. **M. Skotak:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.19/BB3

Topic: C.09. Brain Injury and Trauma

Support: CNRM Grant 308049-14.01-60855

Title: A novel monoacylglycerol lipase inhibitor MJN110 reduces neuroinflammation in the mild repetitive closed head injury mouse model

Authors: *P. SELVARAJ[1], J. WEN[1], M. TANAKA[1], Y. ZHANG[2]
[1]Anatomy, Physiology, and Genet., Henry M. Jackson Foundation/Uniformed Services Uni, Bethesda, MD; [2]Anatomy, Physiology, and Genet., Uniformed Services Univ. of the Hlth. Sci., Bethesda, MD

Abstract: Mild traumatic brain injury (mTBI) is the predominant type of brain injury in young adults and is a risk factor for developing chronic traumatic encephalopathy and other neurodegenerative diseases. Our previous studies have demonstrated that elevation of endocannabinoids 2-arachidononyl glycerol and anandamide by inhibition of α/β-hydrolase domain 6 and fatty acid amide hydrolase reduces neuroinflammation and attenuates neuronal injury in the controlled cortical impact mouse model. Here, we investigate the therapeutic efficacy of MJN110, a novel monoacylglycerol Lipase (MAGL) inhibitor in a repetitive mild closed head injury (rmCHI) mouse model. rmCHI procedure was performed in 8-10 week old, male C57BL/6 mice once a day for 3 days. MJN110 (2.5 mg/kg, i.p.) was administered at 30 min after each impact and then once a day for 5 days. To evaluate fine motor movement and motor coordination, beam walk and rotarod tests were conducted at 4, 6 and 10 days after first impact. Our results showed that treatment with MJN110 significantly reduced foot-faults in beam walk and rendered longer latency to fall in the rotarod test. Learning and memory defects in TBI mouse were observed at day 13 by Y maze and Days 24-28 by Morris water maze, and these deficits were significantly reversed in the drug treated animals. Treatment with MJN110 significantly attenuated the expression of proinflammatory cytokines including IL-6, TNF-α, iNOS and ATF-3 (a stress inducible transcription factor) in ipsilateral cortex and suppressed the accumulation of astrocytes and microglia/macrophages in hippocampus. The increased expression of phosphorylated Tau (p-Tau) in ipsilateral hippocampal dentate gyrus was dramatically reduced. Western blot analysis revealed that MJN110 significantly reduced the increased expressions of amyloid precursor protein (APP), p-Tau and iNOS in the cerebral cortex of TBI mouse brain. MJN110 treatment also restored the expression of the glutamate receptor subunits, GluR1, NMDAR2A and 2B, which showed a significant decrease in the rmCHI mouse cortex and hippocampus. We speculate that the reduced inflammatory response and restored glutamate receptor expression might contribute to the improved motor function, learning and memory in MJN110 treated animals. The therapeutic mechanisms afforded by MJN110 is currently under investigation.

Disclosures: **P. Selvaraj:** None. **J. Wen:** None. **M. Tanaka:** None. **Y. Zhang:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.20/BB4

Topic: C.09. Brain Injury and Trauma

Support: Department of Veterans Affairs (Veterans Health Administration, Office of Research and Development, Rehabilitation Research and Development (I01RX001520)),

Assistant Secretary of Defense for Health Affairs through the Congressionally Directed Gulf War Illness Research Program (W81XWH-16-1-0626)

Florida Department of Health James and Esther King Biomedical Research Program (4KB14)

Bay Pines Foundation

Title: Acute expression changes potentially affecting chronic responses in a mouse mild repetitive TBI model

Authors: *W. A. RATLIFF[1,2], D. C. DRISCOLL[1], S. NEYRA[1], D. QUPTY[3], C. G. PICK[3], B. A. CITRON[1,2]
[1]Bay Pines VA Healthcare Syst., Bay Pines, FL; [2]Dept. of Mol. Med., Univ. of South Florida Morsani Col. of Med., Tampa, FL; [3]Dept. of Anat. and Anthrop., Tel Aviv Univ. Sackler Sch. of Med., Tel Aviv, Israel

Abstract: Worldwide, the incidence of traumatic brain injury (TBI) approximates 0.5% per year and the frequency is much higher among service personnel and athletes. The majority of TBIs are mild and these can result in deleterious cognitive effects for which there is currently no effective treatment. Moreover, repetitive mild head injuries, common in military and athletic activities, have produced more severe consequences. We have demonstrated improved outcomes in both *in vitro* and *in vivo* models of brain injury following treatment with tert-butylhydroquinone (tBHQ) by activation of the inflammatory responsive transcription factor, Nrf2, and downstream neuroprotective factors. Additionally, the PPAR-γ agonist, pioglitazone, has been shown to have neuroprotective effects in models of neurodegenerative disease and TBI. In an effort to better understand the underlying mechanisms we tested mice receiving closed head injuries once per week for 5 weeks along with potentially synergistic treatment by tBHQ plus pioglitazone. At acute and chronic times we evaluated gene expression, cognitive changes, and immunohistochemistry for microglial changes (Iba1) and lipid peroxidation (4-hydroxynonenal). mRNA samples from the ipsilateral hippocampi one day post-injury were evaluated with Affymetrix GeneChip Arrays. Our initial examination (4 groups, n=6 per group) indicated that genes displayed a variety of expression patterns. For example, dysregulations after injuries alone included upregulation of G-protein coupled receptor 3 (Gpr3) and downregulation of aminolevulinic acid synthase (Alas2). After injuries, the transcription factor modulation caused elevation of cysteine-rich secretory protein LCCL domain containing 2 (Crispld2) and lowering of erythroid differentiation regulator 1 (Erdr1) expression. Some genes that were decreased by the injury were increased by the treatment, e.g., SRY (sex determining region Y)-box 3 (Sox3), and certain genes, like ankyrin repeat and SOCS box-containing 6 (Asb6), were induced by the injury and reduced by the treatment. Through these approaches, we hope to better define inflammatory responsive transcription factor signaling pathways and identify factors that could be targeted to produce neuroprotection and improve outcomes for our veterans and others that have received TBIs.

Disclosures: **W.A. Ratliff:** None. **D.C. Driscoll:** None. **S. Neyra:** None. **D. Qupty:** None. **C.G. Pick:** None. **B.A. Citron:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.21/BB5

Topic: C.09. Brain Injury and Trauma

Title: Phenserine mitigates neurodegeneration and neuroinflammation in a mouse mild traumatic brain injury model

Authors: D. LECCA[1], M. BADER[2], D. TWEEDIE[1], R. BECKER[3], C. PICK[2], *N. H. GREIG[1], *N. H. GREIG[1], *N. H. GREIG[1]
[1]Drug Design & Develop. Section, LNS, Intramural Res. Program, Natl. Inst. On Aging, NIH, Baltimore, MD; [2]Tel Aviv Univ., Tel Aviv, Israel; [3]Independent researcher, Park City, UT

Abstract: Mild traumatic brain injury (mTBI), as a single or repeated event, is a common cause of a large spectrum of symptoms, including headache, depression, anxiety, and impaired cognitive function. Associated with neuronal dysfunction and loss, mTBI may promote later development of neurodegenerative disorders. No pharmacological treatment has been approved for TBI. Herein, we evaluated the neuroprotective and anti-inflammatory potential of phenserine (Phen), a reversible anticholinesterase with a broad range of non-cholinergic actions originally developed and clinically tested for Alzheimer's disease, in a mouse mTBI (30 g weight drop) model. By immunohistochemical techniques, we evaluated Phen at 2 clinically translatable doses (2.5 & 5.0 mg/kg BID post mTBI) in hippocampus and lateral parietal cortex (LPC) of mTBI-challenged vs. control (unchallenged) mice at 72 hr (time of peak apoptosis). mTBI induced neuronal loss within all analyzed brain regions, as evaluated by counting Fluorojade C positive (+) cells, which Phen (5.0 mg/kg) fully mitigated. Neuroinflammatory changes were appraised by quantitative evaluation of microglial activation (IBA1) and pro-inflammatory cytokine TNFα immunoreactivity (IR) levels. Following mTBI, IBA1 IR was elevated vs. controls, across all analyzed brain regions. Treatment with Phen (5.0 mg/kg) inhibited microglial activation in CA3 and dentate gyrus (DG) of hippocampus, and in LPC. Consistent with this, IR for the pro-inflammatory cytokine TNFα was elevated in IBA1+ cells in mTBI-challenged vs. control mice across brain areas. Phen (2.5 & 5.0 mg/kg) reduced levels IBA1/TNFα IR colocalization volume in CA3 and DG for hippocampus, and in LPC. These data indicate that Phen ameliorated the neurodegeneration and neuroinflammation mediated by mTBI, consistent with these same doses mitigating cognitive impairments in mice at 7 days (Tweedie et al., PLoS One, 2016); offering a therapeutic approach for this pathological condition.

Disclosures: **D. Lecca:** None. **M. Bader:** None. **D. Tweedie:** None. **R. Becker:** None. **C. Pick:** None. **N.H. Greig:** None. **N.H. Greig:** None. **N.H. Greig:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.22/BB6

Topic: C.09. Brain Injury and Trauma

Support: The Dr. Miriam and Sheldon G. Adelson Chair and Center for the Biology of Addictive Diseases, Tel Aviv, Israel

This research was supported in part by the Ari and Regine Aprijaskis Fund, at Tel-Aviv University. Tel aviv 6920337, Israel.

Title: Hyperbaric oxygen therapy as a treatment for traumatic brain injury

Authors: *C. G. PICK[1], S. TOUSSIA- COHEN[2], R. BARATZ- GOLDSTEIN[2], A. ELPAZ[2], V. RUBOVITCH[2]
[2]Anat., [1]Tel Aviv Univ., Tel Aviv, Israel

Abstract: Traumatic brain injury is the most common cause of death and chronic disability among people under 35-years-old and without an effective pharmacological treatment currently existing. Hyperbaric oxygen therapy (HBOT) is defined as the inhalation of pure oxygen in a hyperbaric chamber that is pressurized greater than 1 atmosphere. HBOT offers physiological and mechanical effects by inducing a state of increased pressure and hyperoxia. HBOT has been proposed as an effective treatment for mTBI, yet the exact therapeutic window and mechanism that underlies this effect is not completely under stood. HBOT was administrated for 4 consecutive days, post a mouse closed head weight drop mild TBI (mTBI) in 2 different time lines: immediate initiated 3 hours post injury and a delayed treatment initiated 7 days post injury. Behavioral cognitive tests and biochemical changes were assessed. The results were similar for both the immediate and the delayed treatments. mTBI mice exhibited impairment in learning abilities, whereas mTBI mice treated with HBO dis played significant improvement compared with the mTBI group, performing similar to the sham groups. mTBI mice had a decline in the myelin basic protein, an increase in neuronal loss (NeuN staining), and an increase in the number of reactive astrocytes (GFAP). The HBO treated mice in both groups did not exhibit these changes and remained similar to the sham group. The delay HBOT has a potential to serve as a neuro protective treatment for mTBI with along therapeutic window.

Disclosures: **C.G. Pick:** None. **S. Toussia- Cohen:** None. **R. Baratz- Goldstein:** None. **A. Elpaz:** None. **V. Rubovitch:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.23/BB7

Topic: C.09. Brain Injury and Trauma

Title: Incretin mimetics as a new drug strategy to treat mild traumatic brain injury

Authors: *M. BADER[1], Y. LI[2], I. TAMARGO[2], D. TWEEDIE[2], V. RUBOVITCH[1], R. D. DIMARCHI[3], K. TALBOT[4], N. H. GREIG[2], C. G. PICK[1]
[1]Dept. of Anatomy, Sackler Fac. of Med., Tel-Aviv Univ., Tel Aviv-Yafo, Israel; [2]Translational Gerontology Branch, Intramural Res. Program, Natl. Inst. on Aging, Natl. Inst. of Hlth., Baltimore, MD; [3]Dept. of Chemistry, Indiana Univ., Bloomington, IN; [4]Dept. of Neurol., UCLA, Los Angeles, CA

Abstract: Traumatic brain injury (TBI) is a commonly occurring injury in victims of sports, motor vehicle accidents and falls. TBI has become a pressing public health concern with no specific therapeutic treatment. Mild TBI (mTBI), which accounts for approximately 90% of all TBI cases, is frequently underestimated, as the immediate physical symptoms decrease rapidly and patients do not show clear structural brain defects. However, they frequently suffer from long-lasting cognitive, behavioral and emotional impairments. The incretins glucagon-like peptide-1 (GLP-1) and glucose-dependent insulinotropic polypeptide (GIP) are gastrointestinal hormones. These peptides induce glucose-dependent insulin secretion, promote β-cell proliferation and enhance resistance to apoptosis. GLP-1 mimetics are on the market as treatments for type II diabetes and are well tolerated. Both GLP-1 and GIP mimetics have shown neuroprotective properties in animal models of Parkinson's and Alzheimer's disease. The aim of this study is to evaluate the potential benefit of enhancing the incretin activity in the brain following mTBI by using two different drugs; liraglutide (GLP-1 analogue) and twincretin (dual GLP-1/GIP receptor agonist). To do that, we subjected mice to mTBI using a weight-drop device and administered twincretin or liraglutide in a 7-day regimen of subcutaneous injection starting 30 min after the injury. Subsequently, we investigated the effects of these drugs on cognitive impairments, neuronal degeneration and neuroinflammation following mTBI over several times. Both liraglutide and twincretin ameliorated mTBI-induced visual memory impairments as assessed by the novel object recognition paradigm, with twincretin showing superiority. Both drugs similarly mitigated mTBI-induced spatial memory deficits in the Y-maze test. These effects were observed at both 7 and 30 days post-mTBI. No differences between groups were evident in anxiety-like behavior assessed by the elevated plus maze demonstrating that efficacy in the preceding paradigms were not confounded by anxiety differences among the groups. Neuronal loss was quantified immunohistochemically using Fluoro-Jade B and anti-NeuN antibodies in the cortex. Both treatments significantly mitigated mTBI-induced neuronal

degeneration. Neuroinflammation was evaluated by the anti-GFAP antibody. mTBI resulted in elevated astrocytes count. No treatment affected this elevation. These findings offer a new potential therapeutic approach to treat mTBI. Further studies will continue to investigate the neuroprotective effects and mechanism of action of incretin-based therapies within the brain.

Disclosures: M. Bader: None. **Y. Li:** None. **I. Tamargo:** None. **D. Tweedie:** None. **V. Rubovitch:** None. **R.D. DiMarchi:** None. **K. Talbot:** None. **N.H. Greig:** None. **C.G. Pick:** None.

Poster

392. Animal Models of Brain Injury: Molecular Mechanisms - Inflammation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 392.24/BB8

Topic: C.09. Brain Injury and Trauma

Title: The GLP-1R agonist, exendin-4, ameliorates open-field blast brain injury-induced learning and memory deficits, neuronal degeneration and altered synaptophysin staining in mouse

Authors: *L. RACHMANY RABER[1], D. TWEEDIE[2], V. RUBOVITCH[1], B. J. HOFFER[3], N. H. GREIG[2], C. G. PICK[1]
[1]Dep.Of Anat. and Anthropology, Sackler Sch. of Medicine, Tel-Aviv Univ., Tel-Aviv, Israel; [2]Intramural Res. Program, Natl. Inst. on Aging, NIH, Baltimore, MD; [3]Case Western Reserve Univ. Sch. of Med., Cleveland, OH

Abstract: Blast Traumatic Brain Injury (B-TBI) is a significant cause of disability and death worldwide, particularly among military forces serving in modern combat operations and civilians. A blast detonation of an explosive device causes changes in the atmospheric pressure, and as a result, organs and tissues of different densities are accelerated at different relative rates, resulting in displacement, stretching and shearing forces. Presently there is no FDA approved medication for the treatment of B-TBI. We investigated the benefits of a FDA approved medicine, Exendin-4 (Exenatide, Ex-4) a long acting glucagon-like peptide 1 receptor agonist, with known neuroprotective/neurotrophic/anti-inflammatory properties in an open field B-TBI model. B-TBI induced long-lasting cognitive and memory impairments. These deficits were associated with B-TBI-dependent diffuse neuronal degenerative process and reductions in the levels of synaptophysin protein staining in cortical and hippocampal tissues. Treatment of animals with a clinically translatable dose of Ex-4 delivered by subcutaneous micro-osmotic pumps to maintain steady-state drug levels for 7 days starting either 48 hours prior to or 2 hours immediately following B-TBI prevented the induction of cognitive and memory deficits as demonstrated in Novel Object Recognition and Y-maze behaviors and the Injury-induced neuronal degeneration and changes in synaptophysin staining. Even very mild exposure to blast wave might cause brain injury. This kind of injury strikes mostly military personnel but also

civilians encountering acts of terrorism and its consequences might be prolonged. Therefore, finding medical solutions to treat the injury is critical and essential. Currently, there is no clinical treatment for blast injury preventing future damage, probably due to the complexity of the brain injury. These data indicate that treatment with the clinically approved drug, Ex-4, represents a viable option for the management of secondary events triggered by blast-induced, mild traumatic brain injury that is commonly observed in militarized areas. Current studies are optimizing Ex-4 administration to support rapid clinical translation.

Disclosures: L. Rachmany Raber: None. **D. Tweedie:** None. **V. Rubovitch:** None. **B.J. Hoffer:** None. **N.H. Greig:** None. **C.G. Pick:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.01/BB9

Topic: C.09. Brain Injury and Trauma

Support: T32 DA007237

NIH/NINDS R01 NS086570-01

The Shriners Hospitals for Children 85110-PHI-14

Title: Loss of blood-brain barrier integrity in the corticostriatal pathway following experimental traumatic brain injury and subsequent susceptibility to the rewarding effects of a sub-threshold dose of cocaine

Authors: *L. CANNELLA[1], M. MAYNARD[1], S. F. MERKEL[1], E. M. LUTTON[1], R. RAZMPOUR[1], R. G. AZER[1], A. M. ANDREWS[3], S. M. RAWLS[2], S. H. RAMIREZ[1]
[1]Pathology and Lab. Med., [2]Ctr. for Substance Abuse Res., Temple Univ. Lewis Katz Sch. of Med., Philadelphia, PA; [3]Pathology and Lab. Med., Temple Univ. L, Philadelphia, PA

Abstract: Substance use disorders are a common de novo psychiatric diagnosis seen in traumatic brain injury (TBI) patients. Previously we found that moderate experimental TBI increased the susceptibility to the rewarding effects of a sub-threshold dose (2.5 mg/kg) of cocaine in adolescent mice two weeks post injury. Interestingly, we did not see this effect if injury occurred during young adulthood or if tested four weeks post injury. Consistent with known secondary mechanisms of injury following TBI, we observed loss of blood-brain-barrier (BBB) integrity and augmented inflammatory profiles, although distal from the area of impact, in brain regions associated with motivation, saliency and reward perception. Specifically, we detected disrupted BBB properties in vessels isolated from the cortex, prefrontal cortex (PFC), and nucleus

accumbens (NAc) of TBI animals measured by decreased expression of tight junction proteins. We also report marked changes in immune response-associated gene expression and increased BBB permeability in these nuclei. Thus, we hypothesize that the BBB changes post brain injury, as a component of neuroinflammation, affect how the rewarding effects of cocaine may shift as a consequence of TBI. An important question that has yet to be answered is whether TBI selectively alters the sensitivity to rewarding effects of psychostimulants or alters responses to their pharmacodynamics, such as induced locomotion. To investigate this we used a controlled cortical impactor set to produce a moderate or mild TBI in 6-week-old male C57BL/6 mice. Locomotor activity was assessed following a single administration of vehicle, 2.5, or 10 mg/kg cocaine in un-injured, sham, mild, or moderate TBI mice two weeks post injury. Ambulation, stereotypy, and total activity were recorded for 60 minutes following acute cocaine exposure. Moderate TBI did not affect locomotor activity produced by cocaine administration. In combination with recent data implying altered expression of dopamine-associated genes in the NAc, our studies suggest that TBI during adolescence may increase the abuse liability of cocaine, as indicated by increased drug-seeking behavior, by altering dopamine neurotransmission in the reward pathway, without increasing cocaine's locomotor activating properties.

Disclosures: L. Cannella: None. **M. Maynard:** None. **S.F. Merkel:** None. **E.M. Lutton:** None. **R. Razmpour:** None. **R.G. Azer:** None. **A.M. Andrews:** None. **S.M. Rawls:** None. **S.H. Ramirez:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.02/BB10

Topic: C.09. Brain Injury and Trauma

Support: NIH Grant R01 NS086570-01

NIH Grant P30 DA013429-16

Shriners Hospitals for Children 85110-PHI-14

Title: Differential changes in tight junction protein expression at areas of blood-brain barrier hyperpermeability following experimental traumatic brain injury

Authors: *M. MAYNARD[1], A. M. ANDREWS[2], E. M. LUTTON[2], R. RAZMPOUR[2], M. SEASOCK[2], W. R. KUNG[2], S. H. RAMIREZ[3]

[1]The Lewis Katz Sch. of Med. at Temple Univer, Philadelphia, PA; [2]Pathology & Lab. Med., [3]Pathology and Lab. Med., The Lewis Katz Sch. of Med. at Temple Univ., Philadelphia, PA

Abstract: Traumatic brain injury (TBI) is an important public health problem in the US. Primary injury from TBI results in physical damage to the neurovascular unit. The secondary phase of injury is triggered by oxidative stress, inflammatory responses and changes in cerebral blood flow. Additionally, it has been well established that dysfunction of the blood-brain barrier (BBB) is a key contributor to the pathology of TBI. However, a biochemical characterization of how tight junction proteins (TJPs), which are essential for BBB integrity, are affected following TBI has not been previously reported. Therefore, using the controlled cortical impact mouse model of TBI (CCI-TBI), vessels were isolated from the site of injury at various time points and across TBI severities. First, visual assessment of BBB permeability was performed using a sodium fluorescein (NaF) in vivo permeability assay. Results demonstrated hyperpermeability to NaF following CCI-TBI, as indicated by increased fluorescent signal in the brain parenchyma of animals that received mild or moderate CCI-TBI at 8, 24 and 72 hrs. Vessels isolated from cortical regions ipsilateral to the impact site were immunostained for the following TJPs: occludin, tricellulin, claudin-5 and ZO-1. Next, vessels were counterstained for endothelial markers PECAM-1 or Glut-1. Image analysis was performed and intensity measurements were calculated as a function of area and vessel caliber. Results demonstrated lower TJP expression following CCI-TBI in the moderate injury groups when compared to sham surgery controls, with distinct protein-specific rates of decreased expression. Morphological changes were also evident, in that some TJPs (i.e. occludin) seemed to be absent from the injured vessels in contrast to others which had a more punctate or cytosolic redistribution. Mild CCI-TBI had a similar profile to moderate for occludin but differed in magnitude. Therefore, our results suggest that various TJPs at the BBB are affected differently by neurotrauma and that ongoing dynamic changes in their expression may offer an explanation to the increased vascular permeability that is observed following TBI.

Disclosures: M. Maynard: None. **A.M. Andrews:** None. **E.M. Lutton:** None. **R. Razmpour:** None. **M. Seasock:** None. **W.R. Kung:** None. **S.H. Ramirez:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.03/BB11

Topic: C.09. Brain Injury and Trauma

Support: Deutsche Forschungsgemeinschaft (SFB 1089)

Fritz Thyssen Foundation

EraNet DeCipher (BMBF)

Local funding (BONFOR)

Title: Intraventricular *In utero*-electroporation BRAFV600E-induced tumors in mice resemble ganglioglioma key features

Authors: *S. CASES-CUNILLERA[1], B. K. ROBENS[2], S. SCHOCH[3], A. J. BECKER[4]
[1]Inst. für Neuropathologie, Universitäts Klinikum Bonn, Bonn, Germany; [2]Univ. Bonn Med. Ctr., Bonn, Germany; [3]Bonn Univ. Med. Sch., Bonn, Germany; [4]Neuropathology; Univ. Bonn Med. Ctr., Bonn, Germany

Abstract: Gangliogliomas (GGs) are generally benign glioneuronal tumors strongly associated with focal epilepsy emerging in younger aged patients. The histological hallmark is an admixture of dysplastic, sometimes bi-nuclear neurons with neoplastic astroglia. Recently, $BRAF^{V600E}$ has been found in approximately 20-60% of human GGs in both, neuronal and astroglial tumor cell components. These data have prompted the concept that GGs can develop by only a 'single mutational' hit to a neural precursor cell population during brain development. To test this hypothesis, we applied intraventricular *in utero* electroporation (IUE) with the piggyBac transposon system to drive genomic integration of transgenes into progenitor cells. We IU-electroporated a piggyBac vector containing the sequence encoding $BRAF^{V600E}$ under control of the ubiquitous (CAG-) promoter at the embryonic day 14 (E14) in CD1/C57BL6 mice (n=5). We observed highly differentiated glioneuronal tumors in all mice subjected to IUE with CAG-$BRAF^{V600E}$ (n=5) vs. no tumors in mice IU-electroporated with CAG-GFP (n=4). Respective tumors are composed of irregularly oriented ganglionic neurons and sparse, isomorphic glial cells with astrocytic differentiation, strongly resembling human GGs. Immunolabeling revealed 48.4% percent of cells within the tumor to be positive for the glial marker Olig2 and 23.4% were positive for the neuronal marker NeuN. These neoplasms showed a low-grade biological behavior with 80% of animals surviving until P110 and, accordingly, antibodies against the proliferation-associated antigen Ki-67 were only detected in less than 5% of cellular nuclei in the area of the tumor. Our data suggest, that the distinct astroglial and dysplastic neuronal components of GGs can develop from a unique mutational hit of neural precursors and that thus, both components can be targeted by a BRAF-inhibitory pharmacological approach.Our work is supported by the Deutsche Forschungsgemeinschaft (SFB 1089), Fritz Thyssen Foundation, the EraNet DeCipher (BMBF) and local funding (BONFOR). We thank Dr. David Jones for providing the DNA encoding $BRAF^{V600E}$.

Disclosures: S. Cases-Cunillera: None. **B.K. Robens:** None. **S. Schoch:** None. **A.J. Becker:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.04/BB12

Topic: C.09. Brain Injury and Trauma

Support: MOMRP, Task area I

Title: Long-term effects of blast exposure: A functional study in rats using an advanced blast simulator

Authors: *P. ARUN, D. M. WILDER, M. A. BENTON, C. M. CHRISTOFOROU, W. DRIWECH, R. T. URIOSTE, O. D. EKEN, V. SAJJA, S. A. VAN ALBERT, Y. WANG, I. D. GIST, J. B. LONG
Ctr. for Military Psychiatry and Neurosci., Walter Reed Army Inst. of Res., Silver Spring, MD

Abstract: Anecdotal observations of blast victims indicate that significant neuropathological and neurobehavioral defects develop at later stages of life. In this study, we have examined the neurobehavioral changes in rats up to 9 months after exposure to single and tightly coupled repeated blasts using an advanced blast simulator. Neurobehavioral changes were monitored at acute (1-15 days), sub-acute (16-30 days) and chronic (1-9 months) time points up to 9 months using elevated plus maze, Y-maze, water maze, open field exploration, rotary pole, rotarod and novel object recognition tests. Single and closely coupled repeated blast exposures resulted in significant functional deficits at both acute and chronic time points. In most functional tests, rats exposed to repeated blasts performed more poorly than rats exposed to single blast. Rotating pole and rotarod tests to assess the neuromotor/balance functions revealed significant deficits at acute and sub-acute time points after blast exposures, and all experimental subjects, including sham controls, had difficulties with these tests at chronic time points. Interestingly, several functional deficits were more pronounced in blast exposed rats at 6 months and beyond. The most substantial changes in blast-exposed rats were observed in the center time and margin time legacies in the open field exploration test at 6 and 9 months after blast exposures. Notably, these two outcome measures were minimally altered acutely, fully recovered during sub-acute stages, and were pronounced during the chronic stages after blast exposures. Significant neuromotor impairment occurred at early stages after blast exposure and the severity increased with number of exposures. Water maze test for spatial learning and memory revealed short-term memory impairments at chronic stages, but not at early time points. The pronounced changes in center time and margin time legacies in the open field exploration test after 6 months post-blast implicate development of depressive-like behavior.

Disclosures: **P. Arun:** None. **D.M. Wilder:** None. **M.A. Benton:** None. **C.M. Christoforou:** None. **W. Driwech:** None. **R.T. Urioste:** None. **O.D. Eken:** None. **V. Sajja:** None. **S.A. Van Albert:** None. **Y. Wang:** None. **I.D. Gist:** None. **J.B. Long:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.05/BB13

Topic: C.09. Brain Injury and Trauma

Title: Omega3-polyunsaturated fatty acids attenuate streptozotocin-induced neurogenesis via BDNF signaling in fat1 transgenic mice

Authors: *G. DO HYEONG[1], T. HWANG[1], S.-A. SHIN[2], J. SHIN[2], J. HONG[2], K. LIM[2], J.-J. KIM[2], D. KIM[2], J. RO[3]
[1]Dept. of Med. Sci., [2]Chungnam Natl. Univ., DAEJEON, Korea, Republic of; [3]LES Corp. Inc., DAEJEON, Korea, Republic of

Abstract: Chronic degenerative brain disease in diabetes, known as ‘diabetic encephalopathy’, is a recognized complication that can occur due to long-standing diabetes in patients. The hippocampus appears vulnerable in diabetic subjects that have a higher risk of stroke, dementia, and cognitive decline. Although the decreased neurogenesis in the dentate gyrus was found streptozotocin (STZ)-induced diabetic mice, the effects of ω3-polyunsaturated fatty acids (PUFA) in diabetics have not been studied yet. Here, we report that high ω3-PUFA could ameliorate the hippocampal degeneration in STZ model, using fat-1 transgenic mouse. When STZ was administrated to wild-type mice in order to induce diabetes, hyperglycemia was well induced after 14 days but not in STZ-treated Fat-1 mice. To examine whether omega-3 could protect from the pyramidal cell loss in the dentate gyrus of hippocampus caused by STZ treatment, we immunostained with Ki67- or DCX antibodies. The number of Ki67- or DCX-reactive cells from STZ-treated wild-type mice was decreased, but not in the STZ-treated fat-1 mice. In addition, the level of expression of Brain-derived neurotrophic factor (BDNF) from hippocampal homogenates in STZ-treated wild-type mice was significantly decreased compared to untreated-wild-type mice. However, BDNF expression in STZ-treated fat-1 mice was comparable to STZ-treated wild-type mice. In conclusion, these findings indicate that PUFA may reduce the loss of proliferation and neuronal progenitors in the dentate gyrus of hippocampus in Type-2 diabetes by STZ.

Disclosures: **G. Do Hyeong:** None. **T. Hwang:** None. **S. Shin:** None. **J. Shin:** None. **J. Hong:** None. **K. Lim:** None. **J. Kim:** None. **D. Kim:** None. **J. Ro:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.06/BB14

Topic: C.09. Brain Injury and Trauma

Support: JSPS KAKENHI Grant 16K21372

Title: Administrations of mesenchymal stem/stromal cells (MSCs) produced beneficial effects in models of traumatic brain injury via prevention of blood brain barrier leakage

Authors: *J. WATANABE, H. OHTAKI, K. YAGURA, K. HONDA, S. ARATA
Showa Univ., Tokyo, Japan

Abstract: Traumatic brain injury (TBI) causes multiple long-term defects including a loss of working memory that is frequently incapacitating. Administrations of mesenchymal stem/stromal cells (MSCs) previously produced beneficial effects in models of TBI as well as other disease models. Intravenous human MSCs decreased neutrophil extravasation, expression of matrix metalloproteinase 9 by endothelial cells and neutrophils, and the subsequent blood brain barrier leakage. In this time, the expression of GFAP was significantly increased. When MSCs were co-cultured with scratch wounded astrocytes in vitro, the wound was disclosed much earlier than control. The data suggested that administration of MSCs may activate astrocytes, and be an effective therapy for TBI via prevention of blood brain barrier leakage.

Disclosures: J. Watanabe: None. **H. Ohtaki:** None. **K. Yagura:** None. **K. Honda:** None. **S. Arata:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.07/BB15

Topic: C.09. Brain Injury and Trauma

Support: KAKENHI Grant JP15H02718

AIST

Title: Focal brain lesions induced by ultraviolet irradiation

Authors: *M. NAKATA[1,2], K. NAGASAKA[1,2,3], I. TAKASHIMA[1,3], S. YAMAMOTO[1]
[1]AIST, Tsukuba, Japan; [2]JSPS Res. fellow, Tokyo, Japan; [3]Univ. of Tsukuba, Tsukuba, Japan

Abstract: Animal models of focal brain lesions are produced using electric current, heat, or drugs, including ibotenic acid. Focal brain injury can also be induced by mechanical impact (e.g., fluid percussion). However, breaks in the dura mater can occur at injection or impact sites, inducing cerebrospinal fluid leakage and increasing the risk of infection during long-term experimental periods. Furthermore, these manipulations may also induce cell death in untargeted brain regions due to the propagation of excess excitation from neuronal cell death within the targeted site (excitotoxicity). To avoid these artifacts, we developed a new method to induce focal brain lesions using irradiation with an ultraviolet (UV) ray over intact dura mater. Adult Wistar rats were anesthetized with isoflurane and the skull was removed. UV irradiation was conducted through an optical fiber connected to a UV (UV-A; wavelength 365 nm, about 1.1 mWh) LED light source. A fiber tip (flat circle, 400 μm in diameter) was stereotaxically placed on the targeted brain site (anterior parietal cortex; Bregma -3.7 mm, 2.0 mm from the midline) and contacted the dura mater. Five days after the irradiation, the animals were perfused for histological analysis. Nissl staining revealed that the UV ray injured the cortical surface of the irradiated site. The absence of neuronal marker (NeuN)-immunopositive cells within the lesion core indicated UV-induced depletion of neuronal cells. After 120 min of irradiation with an about 0.55-mW UV ray, we observed a column-shaped lesion core of approximately 600 μm in diameter and 700 μm in depth. In contrast, irradiation with reduced light intensity produced a relatively superficial lesion. Taken together, we succeeded in developing a novel method to create focal brain lesions using UV irradiation. Moreover, our results suggest that this UV-lesion method enables the control of the lesion depth using the light intensity while keeping the dura mater intact.

Disclosures: **M. Nakata:** None. **K. Nagasaka:** None. **I. Takashima:** None. **S. Yamamoto:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.08/BB16

Topic: C.09. Brain Injury and Trauma

Support: Howard Hughes Medical Institute - Exrop 2016

Virginia Commonwealth University Initiative for Maximizing Student Development (VCU IMSD) through the grant number R25 GM090084 (NIH/NIGMS)

Title: Assessing dendritic identity and protein trafficking in regenerated dendrites in adult *Drosophila*

Authors: *S. IZABEL[1,2], Y. JAN[3], L. JAN[3], L. DEVAULT[4]
[1]Anat. and Neurobio., Richmond, VA; [2]Physiol., [3]Physiology, Biochem., [4]Neurosci., Univ. of California San Francisco, San Francisco, CA

Abstract: Neurons exchange information through synaptic signals or sensory inputs received by the dendrites, which may generate action potentials that can propagate along the axon. This exchange can be interrupted when axons or dendrites are damaged. Axonal regeneration has been widely studied, but dendritic regeneration has not received the same attention. For that reason we have focused this study on assessing injury recovery in Drosophila dendrites. The model used has several desirable features: neurons that are easily imaged, a tractable genetic system, and the potential to build a framework for future studies. Previous research has assessed dendritic growth in response to acute injury using Drosophila larvae. We aim to evaluate such growth in adult animals. Drosophila larvae are still developing. We aim to understand how stable adult dendritic arbors respond to injury. When injuring the animals, we utilize lasers from a two-photon microscope to severe all dendrites at the first branch point by laser ablation, leaving a bald neuron. In assessing injury response, we image animals and perform immunohistochemistry staining. The staining assesses the injured neuron's maintenance of neuronal identity, ability to correctly traffic somatodendritic proteins and integrity of cytoskeletal structure. We stained these neurons using the antibody against PPK-26, a neuronal type specific marker, to assess the identity of injured neurons. We also stain for PPK-26 in a non-permealized condition to attest that these proteins are located at the surface of the neuron, as in uninjured neurons. Finally, we test for stabilized microtubules, by staining for Futsch, the Drosophila MAP1B homolog. Preliminary results indicate the presence of PPK-26 in regenerated neurons suggesting that the dendrites present morphological similarities to those of fully functioning healthy neurons. We have also located presence of Futsch marker in regrown dendrites. Although certainty is only reached after quantitative analyses, we hope to find that these dendrites also contain microtubules in its regenerated expansions. Ultimately, we aspire to expand our knowledge of the cytoskeletal structure of regenerated dendrites and compare with findings of the larvae model, which have established that regrown dendrites maintain some but not all of the characteristics of uninjured dendrites.
The Howard Hughes Medical Institute and Exrop Research Training Program supported this work

Disclosures: **S. Izabel:** Other; Howard Hughes Medical Institute, National Institute of Health. **Y. Jan:** None. **L. Jan:** None. **L. DeVault:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.09/BB17

Topic: C.09. Brain Injury and Trauma

Support: NRF-2017R1C1B1004226

Title: Acamprosate reduces traumatic brain injury-induced neuronal death in rats

Authors: *B. CHOI[1], S. LEE[1], S. MIN[2], T. CHUNG[2], H. CHOI[3], S. SUH[1]
[1]Dept. of Physiol., Hallym University, Col. of Med., Chuncheon, Korea, Republic of; [2]Dept. of Emergency Med., CHA Bundang Med. Center, CHA University, Sch. of Med., Seongnam, Korea, Republic of; [3]Dept. of Neurol., Hallym University, Collge of Med., Chuncheon, Korea, Republic of

Abstract: Acamprosate, or N-acetyl homotaurine, is a N-methyl-D-aspartate (NMDA) receptor antagonist that is used as a pharmacological means of treatment for chronic alcohol dependence. Although the exact mechanism of acamprosate has not been clearly established, it appears to work by promoting a balance between the excitatory and inhibitory neurotransmitters, glutamate and gamma-aminobutyric acid (GABA), respectively. Several studies have demonstrated that acamprosate provides neuroprotection against cerebral ischemia-induced brain injury. However, there are no studies investigting the role of acamprosate on traumatic brain injury (TBI)-induced neuronal death. In the present study, we sought to analyze the therapeutic potential of acamprosate to protect against neuronal death and other underlying pathogenic mechanisms that arise following TBI. Rats were given acamprosate (200 mg/kg) orally once per day for two weeks. Two week later, rats were subjected to a controlled cortical impact (CCI; 5 m/sec, 500-msec duration, 5-mm deformation) injury over the right parietal cortex. Histological analysis was performed at 3 or 24 hours, or 7 days after TBI. We found that acamprosate treatment for 2 weeks reduces levels of vesicular glutamate and zinc in the hippocampus. Consequently, this reduced vesicular glutamate and zinc level resulted in reduction of ROS production at 3 hours after TBI. When evaluated at 24 hours after TBI, acamprosate administration reduced the number of degenerating neurons, blood-brain barrier (BBB) disruption, leukocyte infiltration and dendritic loss. Acamprosate also reduced glial activation and neuronal loss at 7 days after TBI. The present study demonstrates that acamprosate attenuates TBI-induced brain damage by depletion of vesicular glutamate and zinc levels. Therefore, the present study suggests that acamprosate may have a high therapeutic potential for prevention of TBI-induced neuronal death.

Disclosures: **B. Choi:** None. **S. Lee:** None. **S. Min:** None. **T. Chung:** None. **H. Choi:** None. **S. Suh:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.10/BB18

Topic: C.09. Brain Injury and Trauma

Support: HRF-201701-017

HRF-S-53

Title: Protective effects of dichloroacetic acid on traumatic brain injury induced neuronal death

Authors: *S. LEE, B. CHOI, S. SUH
Dept. of physiology, Col. of Med., Chunchen, Korea, Republic of

Abstract: Traumatic brain injury (TBI) is defined as damage to the brain resulting from external mechanical force, such as rapid acceleration or deceleration, impact, blast waves, or penetration by a projectile. A penetrating or open head injury occurs when an object pierces the skull and breaches the dura mater, the outermost membrane surrounding the brain. Neuronal death occurs in the hippocampus due to this traumatic brain injury. Dichloroacetic acid (DCA) is a small molecule that has multiple effects on intermediary metabolism. Biochemically, DCA inhibits gluconeogenesis, lipogenesis and cholesterol synthesis and stimulates peripheral glucose and lactate utilization. DCA has also been studied as a potential drug due to its ability to inhibit the enzyme pyruvate dehydrogenase kinase. The present study aimed to evaluate the therapeutic potential of DCA on TBI-induced neuronal death. Traumatic brain injury was performed using a weight drop model in rat. DCA (100mg/kg) was injected into the intraperitoneal space immediately after TBI. Neuronal death was evaluated with Fluoro Jade-B (FJB) staining 24h after TBI. DCA reduces the number of degenerating neurons in CA3 and DG of hippocampus after TBI. Oxidative injury was detected by 4-hydroxy-2-nonenal (4HNE) and microglia activation was detected by CD11b immunohistochemistry in the hippocampus after TBI. DCA treatment decreased the intensity of 4HNE fluorescence and microglia activation in the hippocampus compared to the saline-treated group after TBI. Therefore, the present study suggests that DCA may have protective effects on traumatic brain injury-induced hippocampal neuronal death. Keywords: Traumatic brain injury, Dichloroacetic acid, Oxidative injury, Microglia

Disclosures: **S. Lee:** None. **B. Choi:** None. **S. Suh:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.11/BB19

Topic: C.09. Brain Injury and Trauma

Support: HRF-S-53

Title: Effects of combined treatment of dichloroacetic acid (DCA) with pyruvate on global cerebral ischemia-induced hippocampal neuronal death

Authors: *D. HONG[1], A. KHO[1], S. SUH[1], M. CHOI[2], O. RYU[2], M. SOHN[3]
[2]Endocrinol. and Metabolism, [1]Hallym Univ., Chuncheon, Korea, Republic of; [3]Nursing, Inha Univ., Incheon, Korea, Republic of

Abstract: Global cerebral ischemia (GCI) occurs when blood flow toward the brain is blocked or dramatically reduced. In this condition, oxygen and glucose provide into the brain was decreased. GCI induces vesicular zinc release, oxidative injury, neuronal death and cognitive impairment. Dichloroacetic acid (DCA) inhibits pyruvate dehydrogenase kinase (PDK), which increases the uptake of pyruvate into the mitochondria and promotes glucose oxidation during glycolysis through enhancement of pyruvate dehydrogenase (PDH) activity. Here, we tested our hypothesis that DCA can prevent neuronal damage after GCI by inhibition of PDK activity since DCA increases the rate of pyruvate converted to ATP. Global cerebral ischemia (GCI) was induced by occlusion of both common carotid arteries (CCAO) and blood was withdrawn through the femoral artery to induce a 7 minute isoelectric period, evaluated by monitoring EEG changes. Eight week old male Sprague-Dawley rats were used and neuronal death was evaluated with Fluoro Jade-B (FJB) staining at 7 days after GCI. To test the therapeutic potential of DCA, rats were given an intraperitoneal injection of DCA (100mg/kg) once per day for 2 days after ischemia. However, there was no difference in the number of FJB + neurons between vehicle and DCA administrated groups after ischemia. Additionally, 50mg/kg of pyruvate was also not neuroprotective after ischemia. Thus, we administrated DCA with pyruvate once per day for 2 days after ischemia. The present study found that administration of DCA with pyruvate significantly decreased neuronal death, oxidative stress, microglia activation, zinc accumulation after GCI. These findings suggest that co-treatment with DCA and pyruvate may increase the regenerative capacity of the hippocampus after stroke or myocardial infarction. Keywords : Global cerebral ischemia, Dichloroacetic acid, Pyruvate, Neuronal death

Disclosures: D. Hong: None. **A. Kho:** None. **S. Suh:** None. **M. Choi:** None. **O. Ryu:** None. **M. Sohn:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.12/BB20

Topic: C.09. Brain Injury and Trauma

Support: HRF-201701-017

HRF-S-53

Title: Effects of sodium dichloroacetate (DCA) on hypoglycemia-induced hippocampal neuronal death

Authors: *A. KHO[1], B. CHOI[1], S. SUH[1], H. CHOI[2], M.-G. CHOI[3], O. RYU[3], T. CHUNG[4]
[1]Dept. of physiology, [2]Neurol., [3]Endocrinol. and Metabolism, Hallym Univ., Chuncheon, Korea, Republic of; [4]Dept. of Emergency Med., CHA Univ. Sch. of Med., Seongnam-si, Gyeonggi-Do, Korea, Republic of

Abstract: Type 1 diabetic patients who are treated with insulin for tight control of blood glucose levels frequently experience severe hypoglycemia when plasma glucose level falls below ~3.5 mM (~63mg/dL), which can lead to seizures and loss of consciousness. Our lab has shown that neuronal cell death caused by hypoglycemia is not only a result of reduced glucose supply to the brain but also proceeds via a cell death process that is initiated by the reperfusion of glucose after a sustained period of glucose deprivation. Severe and prolonged hypoglycemia causes zinc release, oxidative stress, neuronal death and cognitive impairment. Zinc release contributes to neuronal death under severe disease conditions, such as prolonged seizure, ischemia and brain trauma. In addition, zinc can inhibit the key glycolytic enzyme glyceraldehyde-3-phosphate dehydrogenase (GAPDH), making glycolysis not to work properly, which compromised neuronal cells eventually. Sodium dichloroacetate (DCA) is a pyruvate dehydrogenase kinase (PDHK) inhibitor and thus activates pyruvate dehydrogenase (PDH), the gatekeeper of glucose oxidation. The principal site of action of DCA is the pyruvate dehydrogenase (PDH) complex inhibiting PDHK, keeping PDH in the unphosphorylated catalytically active form and replacing pyruvate with acetyl-CoA production, which enhances glucose oxidation in cells, leading to increased levels of ATP. Additionally, DCA has been shown to prevent ROS generation. We hypothesized that DCA treatment can reduce neuronal cell death through improvement of glycolysis and prevention of ROS production after hypoglycemia. To test this, we used an animal model of insulin-induced hypoglycemia and injected DCA (100mg/kg, *i.v*, 2 days) following hypoglycemic insult. Brain sections were stained with Fluoro Jade-B (FJB) to detect degenerating neurons in the hippocampus at 1 week after hypoglycemia. As a result, DCA treatment after hypoglycemia reduced the number of FJB (+) cells compared to hypoglycemia-

vehicle group. Brain sections were also evaluated for oxidative injury using 4-hydroxy-2-nonenal (4HNE) and for microglia activation using CD11b antibody. DCA treatment also significantly reduced hypoglycemia-induced oxidative injury and microglial activation. Therefore, the present study suggests that DCA may have high therapeutic potential to reduce hippocampal neuronal death after hypoglycemia. **Key word**: Hypoglycemia, Sodium dicholoroacetate (DCA), PDH, PDHK inhibitor, Pyruvate, Glycolysis, Oxidative stress, Neuron death

Disclosures: **A. Kho:** None. **B. Choi:** None. **S. Suh:** None. **H. Choi:** None. **M. Choi:** None. **O. Ryu:** None. **T. Chung:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.13/BB21

Topic: C.09. Brain Injury and Trauma

Support: NIH/NIAAA T32AA013527

NIH/NIAAA-R21 AA020951

Title: The effects of alcohol and traumatic brain injury on neural stem cell responses

Authors: *S. T. TON[1], S.-Y. TSAI[2], H. M. FLINK[2], J. Y.-T. WU[2], J. J. HSU[2], I. C. VAAGENES[2], G. L. KARTJE[2]
[1]Neurosci., Loyola, Maywood, IL; [2]Edward Hines Junior VA Hosp., Hines, IL

Abstract: Our laboratory has previously found that a repeated dose of binge alcohol prior to TBI leads to worse recovery on a sensitive test of skilled forelimb function (Vaagenes et al. 2015). Mobilization of endogenous neural stem cells might be one mechanism that contributes to the functional compensation after TBI. We therefore sought to determine the effect of binge alcohol at the time of TBI on subventricular zone (SVZ) neural stem cell responses. Male rats (2 month old Sprague Dawley) underwent binge alcohol (3g/kg/day) by gastric gavage for 2 days prior to TBI. On the day of TBI, rats were given alcohol 1 hour prior to injury using the controlled cortical impact (CCI) method directed to the forelimb sensorimotor cortical area. We assessed the SVZ neural stem cell response after binge alcohol and TBI by utilizing the proliferation marker 5-bromo-2'-deoxyuridine (BrdU) along with other markers for neurogenesis such as Doublecortin. We found that binge alcohol did not affect short and long term lesion size (as measured at 24 hours, 7 days and 6 weeks post TBI). As expected, TBI alone significantly increased SVZ proliferation bilaterally 24 hours post injury. Surprisingly, binge alcohol alone also significantly increased proliferation bilaterally in the SVZ at the 24 hour time point. A

combined binge alcohol and TBI regimen resulted in decreased SVZ proliferation bilaterally at 24 hours and 7 days post-TBI. Furthermore, when assessed at 6 weeks after TBI, binge alcohol significantly decreased SVZ neuronal differentiation. While we observed that TBI alone increased migration toward the rostral migratory stream (RMS), binge alcohol did not affect either RMS or perilesional migration post-TBI. Taken together, these results suggest that TBI combined with binge alcohol decreases short-term neural stem cell proliferative responses as well as long-term neuronal differentiation of these cells in the SVZ, thereby negatively impacting functional recovery. We are currently examining the possible functional implications of the stimulatory effect of binge alcohol alone on SVZ neurogenesis, specifically its connection to olfactory bulb function.
This work was supported by the Loyola University Neuroscience Division, the Department of Veterans Affairs, NIH/NIAAA T32AA013527 and NIH/NIAAA-R21 AA020951.

Disclosures: S.T. Ton: None. **S. Tsai:** None. **H.M. Flink:** None. **J.Y. Wu:** None. **J.J. Hsu:** None. **I.C. Vaagenes:** None. **G.L. Kartje:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.14/BB22

Topic: C.09. Brain Injury and Trauma

Support: Deanship of Research at Jordan University of Science and Technology, Irbid, Jordan

Title: Evaluating the protective effect of etazolate on anxiety- and depression- like behavior and cognitive impairment induced by post traumatic stress disorder

Authors: *K. H. ALZOUBI[1], Z. AL SUBEH[2], O. F. KHABOUR[2]
[1]Jordan Univ. of Sci. & Technol., Irbid, Jordan; [2]Jordan Univ. of Sci. and Technol., Irbid, Jordan

Abstract: Post-traumatic stress disorder (PTSD) occurs after experiencing traumatic events. PTSD symptoms usually include intrusive recollections of traumatic event, avoidance of event reminders, and hyperarousal. The absence of optimal treatment along with low response rate make the investigation for novel pharmacological therapy highly required. Etazolate (ETZ), on the other hand, is a selective PDE-4 inhibitor that is proven to be of particular importance in neuropharmacology. In this study, we aimed to evaluate the possible beneficial effects of etazolate in preventing and reversing stress induced anxiety, depression, and cognitive impairment. Single prolonged stress (SPS) is an animal model of PTSD that can effectively manifests its characteristic symptoms of anxiety, depression, and memory impairment. Current results revealed that ETZ significantly reduce anxiety, depression-like behavior, and memory

impairment induced by PTSD. It also restored oxidative stress biomarkers, and reduced BDNF level, which are suggested as possible molecular targets of ETZ protective effect during PTSD.

Disclosures: K.H. Alzoubi: None. **Z. Al Subeh:** None. **O.F. Khabour:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.15/BB23

Topic: C.09. Brain Injury and Trauma

Support: NINDS 5R21NS096554

Title: Positive allosteric modulation of cholinergic receptors improves spatial learning and memory following brain trauma in mice

Authors: *D. P. HOLSCHNEIDER[1], Y. GUO[1], O. U. SCREMIN[2], Z. WANG[1]
[1]Psychiatry & the Behavioral Sci., USC, Los Angeles, CA; [2]Greater Los Angeles VA Healthcare Syst., Los Angeles, CA

Abstract: AIM: There are currently no clinically efficacious drug therapies to treat neuronal damage following traumatic brain injury. This study examines the potential of a novel cholinergic treatment on improving memory and motor dysfunction with a lower incidence of undesirable side effects that have limited prior pharmacotherapies. **METHODS:** Adult mice received unilateral cortex contusion injury (CCI) or sham injury. Two days thereafter, they were administered benzyl quinolone carboxylic acid (BQCA, 5, 10, 20 mg/kg, i.p.), a M1 muscarinic positive allosteric modulator or vehicle, with dosing twice per day over 3 ½ weeks, and the 1st daily dose given 30 minutes prior to behavioral testing. One week following CCI, mice were evaluated weekly for 3 weeks with a battery of motor tests, as well as with daily evaluation of spatial learning and memory using the Morris Water Maze. **RESULTS:** Unilateral CCI resulted in a significant, persistent, asymmetric postural reflex on tail suspension. Motor testing revealed significant deficits on the accelerating rotarod, increased footslips on grid walking and decreased grip strength and stride length ($p<0.05$), as well as progressive hyperactivity that was significant 3 weeks after CCI ($p<0.05$). Compared to sham-vehicle controls, CCI-vehicle mice demonstrated significantly delayed spatial learning at 2 and 3 weeks post-CCI during exposure to a reverse learning task in which the platform position was moved ($p<0.05$). In CCI mice, BQCA resulted in significantly improved spatial learning and recall at weeks 2 and 3. Effects of BQCA on motor outcomes were minimal. There was a trend toward the highest dose of BQCA attenuating the locomotor hyperactivity seen in CCI animals. BQCA at 20 mg/kg had no significant effect on general motor activity, body weight or acute motor, secretory, or respiratory symptoms.

CONCLUSION: BQCA in CCI mice elicited improved hippocampal-dependent learning with repeated daily exposure to visual cues in a spatial navigation task. Improvements were noted in the absence of significant cholinergic side effects. No significant drug effect was noted on motor outcomes. These results suggest that BQCA is a candidate compound to improve learning and memory function following brain trauma, and may not suffer the associated CNS side effects typically associated with even modest doses of other cholinergic enhancers. Ongoing work is applying functional brain mapping to examine functional reorganization of disrupted neuronal networks in this rodent model of focal brain trauma. **Acknowledgements**: This work was supported by a grant from NINDS 5R21NS096554. Dr. Scremin passed away during completion of this work.

Disclosures: D.P. Holschneider: None. **Y. Guo:** None. **O.U. Scremin:** None. **Z. Wang:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.16/BB24

Topic: C.09. Brain Injury and Trauma

Support: NCTR Protocol E7584.11

Title: Moderate traumatic brain injury induces an increase in blood-brain-barrier permeability and decreases tight junction protein expression

Authors: *S. F. ALI[1], H. ROSAS-HERNANDEZ[2], E. CUEVAS[2], C. ESCUDERO-LOURDES[3], N. GOMEZ-CRISOSTOMO[4], S. M. LANTZ[5], N. STURDIVANT[6], P. RAVISHANKAR[6], K. BALACHANDRAN[6], W. SLIKKER, Jr[7], M. G. PAULE[8]
[1]Neurochemistry Lab, Div. of Neurotoxicology, Natl. Ctr. Toxicological Res/Fda, Jefferson, AR; [2]Natl. Ctr. For Toxicological Res., Jefferson, AR; [3]Facultad de Ciencias Químicas, UASLP, San Luis Potosi, Mexico; [4]UNIVERSIDAD JUAREZ AUTONOMA DE TABASCO (DIVISION A, COMALCALCO, TABASCO, Mexico; [5]Div. of Neurotoxicology, NCTR/FDA, Jefferson, AR; [6]Univ. of Arkansas, Fayetteville, AR; [7]Natl. Ctr. for Toxicological Res., Jefferson, AR; [8]Div. of Neurotoxicology, FDA's Natl. Ctr. For Toxicological Res., Jefferson, AR

Abstract: Traumatic brain injury (TBI) remains one of the major causes of death and disability. Due to the heterogeneity of its causes, the events that occur after TBI can be quite varied and are not well understood. Regardless of cause, deformation of brain tissue can lead to neuronal, glial and endothelial cell death as well as other cellular and molecular responses including changes in blood-brain barrier (BBB) permeability. Understanding the events that occur after TBI is important for the development of therapeutic approaches. The aim of this study is to characterize

the effects of mild and moderate TBI on the BBB in mice using a weight drop method. Mice were anesthetized using isoflurane and placed on a soft foam pad. An acrylic tube was placed directly above the head of the mouse and a 50 gram weight was dropped onto the mouse head from either a 30cm (mild) or 120cm (moderate) height within the tube. 24 hours after a single such TBI episode BBB permeability was evaluated using the Evans blue method and by quantifying the plasma concentrations of the glial-specific protein S100β. The expression of the tight junction proteins zonula occludens-1 (ZO-1) and occludin was evaluated using western blot. Mild TBI did not affect Evans blue extravasation or S100β concentration and did not change the expression of the tight junction proteins; however, moderate TBI significantly increased Evans blue extravasation and the plasma concentrations of S100β, indicating an increase in BBB permeability. This effect was related to a decrease in the expression of ZO-1 and occludin. In summary, we demonstrated using a weight drop model that moderate TBI increases BBB permeability that is likely due to a decrease in the expression of tight junction proteins. This model can be used as a tool to test potential treatments that can protect the BBB after TBI.

Disclosures: S.F. Ali: None. **H. Rosas-Hernandez:** None. **E. Cuevas:** None. **C. Escudero-Lourdes:** None. **N. Gomez-Crisostomo:** None. **S.M. Lantz:** None. **N. Sturdivant:** None. **P. Ravishankar:** None. **K. Balachandran:** None. **W. Slikker:** None. **M.G. Paule:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.17/BB25

Topic: C.09. Brain Injury and Trauma

Support: MEXT Grants-in-Aid for Scientific Research No. 24590133

MEXT Grants-in-Aid for Scientific Research No. 15K07910

MEXT Grants-in-Aid for Scientific Research No. 26462161

Tokushima Bunri University No. TBU2012-2-2

Title: Traumatic brain injury accelerates amyloid-beta deposition and impairs learning and memory in the triple-transgenic mouse model of Alzheimer’s disease

Authors: *Y. KIRINO[1,2], *Y. KIRINO[5], H. SHISHIDO[6], N. KAWAI[8], Y. TOYOTA[6], T. TAMIYA[6], M. UENO[7], T. KUBOTA[3], Y. KISHIMOTO[4]
[2]Sch. of Pharmaceut. Sci., [1]Tokushima Bunri Univ., Tokushima, Japan; [3]Kagawa Sch. of Pharmaceut. Sci., Tokushima Bunri Univ., Sanuki, Kagawa-ken, Japan; [4]Kagawa Sch. of

Pharmaceut. Sci., Tokushima Bunri Univ., Sanuki,Kagawa-ken, Japan; [5]Layton Biosci. Inc, Tokushima, Japan; [6]Dept. of Neurolog. Surgery, Fac. of Med., [7]Dept. of Inflammation Pathology, Fac. of Med., Kagawa Univ., Miki-cho, Japan; [8]Dept. of Neurolog. Surgery, Kagawa Gen. Rehabil. Hosp., Takamatsu, Japan

Abstract: Object : There are many studies that demonstrate correlation between traumatic brain injury (TBI) and Alzheimer's disease (AD). Several large epidemiological examinations have revealed that TBI is an epigenetic risk factor for developing AD. But it is not known how TBI contributes to the onset or progression of this late life dementia. To investigate this question, we studied neuropathological and behavioral consequences of TBI in a triple transgenic AD-model mouse (3×Tg mice) that harboring $PS1_{M146V}$, APP_{SWE}, and tau_{P301L} transgenes. Here, we report that weight drop method TBI in 3×Tg mice demonstrated Aβ deposition and APP accumulation at 7days or 4 weeks after TBI. **Material and method :** Five to seven-month-old 3×Tg mice and wild type mice were subjected to TBI (n = 7-10) or sham treatment (n = 7-8). At 7 days and 4weeks after treatment, we assessed Aβ deposition and APP accumulation in the hippocampus of these mice. And a subset of mice also was studied behaviorally at 7 days and 4 weeks after injury. **Result :** Compared to sham-treatment wild type mice, brain-injured wild type mice exhibited significant deficits in spatial learning ability as measured using the Morris water maze over the first 7 days ($p = 0.018$), but not after 4 weeks post-injury. On the other hand, compared to sham-treatment 3xTg mice, TBI-treated 3×Tg mice demonstrated significant deficits in spatial learning ($p = 0.048$) and slower acquisition of delay eyeblink conditioning ($p = 0.023$) after 4 weeks post-injury accompanied increased Aβ deposition in the hippocampus ($p = 0.018$). **Conclusion :** This is the experimental evidence linking TBI to mechanism of AD by showing that TBI accelerates brain Aβ deposition and cognitive impairment. These data suggested that despite recovery from acute cognitive deficits, concussive brain trauma leads to axonal damage with APP accumulation and increased Aβ disposition induced cognitive impairment after brain injury.

Disclosures: Y. Kirino: None. **Y. Kirino:** None. **H. Shishido:** None. **N. Kawai:** None. **Y. Toyota:** None. **T. Tamiya:** None. **M. Ueno:** None. **T. Kubota:** None. **Y. Kishimoto:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.18/BB26

Topic: C.09. Brain Injury and Trauma

Support: NHMRC

Title: Molecular basis for amyloid precursor protein mediated neuroprotection in brain injury

Authors: *C. L. INAMPUDI[1], L. Q. LAM[2], B. ROBERTS[3], S. PLUMMER[4], F. CORRIGAN[4], G. D. CICCOTOSTO[1], P. J. CRACK[2], C. VAN DEN HEUVEL[4], R. CAPPAI[1]
[1]Dept. of Pathology, The Univ. of Melbourne, Melbourne, Australia; [2]Dept. of Pharmacol. and Therapeut., Univ. of Melbourne, Melbourne, Australia; [3]The Univ. of Melbourne, The Florey Inst. of Neurosci. and Mental Hlth., Melbourne, Australia; [4]Med. Sci., The Univ. of Adelaide, Adelaide, Australia

Abstract: Amyloid precursor protein (APP) is neuroprotective in traumatic brain injury (TBI). Treatment with soluble amyloid precursor protein (sAPP) can rescue motor and cognitive deficits following TBI in mouse and rat models (Corrigan, Vink et al. 2012). The neuroprotective active site in sAPP is located in residues 96 to 100 (APP96-110) (Corrigan 2014). We hypothesize that APP96-110 interacts with specific molecule(s) to trigger its neuroprotective response in TBI. To identify protein(s) interacting with APP96-110 peptide, a biotin-streptavidin affinity capture method followed by mass spectrometry was utilised. Among the proteins identified the Amyloid Precursor-like Protein 2 (APLP2) was found to be a robust interacting target with APP96-110. Previous reports showed APLP2 binds to region in APP which includes 96-110 (Soba, Eggert et al. 2005). To test the neuroprotective interaction between APP and APLP2, APLP2 wild type (APLP2 WT) and APLP2 knockout (APLP2 KO) mice (male and female) have been subjected to TBI and gait parameters assessed by Digigait. Preliminary data shows that APLP2 KO mice are more susceptible to TBI than APLP2 WT mice after 24h of injury. This indicates that the APLP2 expression modulates the rate of brain injury following TBI. The role of APLP2 in mediating APP96-110 neuroprotection is currently being investigated.
References:
Corrigan, F., R. Vink, P. C. Blumbergs, C. L. Masters, R. Cappai and C. van den Heuvel (2012). "sAPPalpha rescues deficits in amyloid precursor protein knockout mice following focal traumatic brain injury." J Neurochem 122(1): 208-220.
Corrigan F, Thornton E, Roisman LC, Leonard AV, Vink R, Blumbergs PC, van den Heuvel C, Cappai R (2014). "The neuroprotective activity of the amyloid precursor protein against traumatic brain injury is mediated via the heparin binding site in residues 96-110". J Neurochem. 28(1):196-204
Soba, P., S. Eggert, K. Wagner, H. Zentgraf, K. Siehl, S. Kreger, A. Löwer, A. Langer, G. Merdes and R. Paro (2005). "Homo-and heterodimerization of APP family members promotes intercellular adhesion." The EMBO journal 24(20): 3624-3634.

Disclosures: C.L. Inampudi: None. **L.Q. Lam:** None. **B. Roberts:** None. **S. Plummer:** None. **F. Corrigan:** None. **G.D. Ciccotosto:** None. **P.J. Crack:** None. **C. Van Den Heuvel:** None. **R. Cappai:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.19/BB27

Topic: C.09. Brain Injury and Trauma

Support: Centre for international Mobility (CIMO)

Finnish Epilepsy Association

Academy of Finland

FP7-HEALTH project 602102 (EPITARGET)

Title: Circulating miR-124 as a predictive biomarker for characterizing post-TBI cortical lesion endophenotypes

Authors: *S. DAS GUPTA, N. VUOKILA, N. PUHAKKA, A. PITKÄNEN
Neurobio., Univ. of Eastern Finland/AIVI, Kuopio, Finland

Abstract: Traumatic brain injury (TBI) encompasses primary brain damage inflicted immediately at the time of impact, and secondary damage involving cellular and molecular alterations progressing over a prolonged time post-trauma. Early diagnosis is thus crucial to identify patients at risk of developing the long-term comorbidities. In this study, we hypothesize that level of the brain-enriched microRNA-124 (miR-124) in circulation may serve as a TBI biomarker, and can predict the chronic cortical lesion endophenotype developing in response to TBI.

For this, we first assessed miR-124 level from plasma, in rats receiving the experimental lateral fluid-percussion injury. Analysis was done with droplet digital PCR (ddPCR). Next, from a cohort of 22 TBI rats with T2-w MRI imaging available at 2mo post-TBI, we characterized the post-TBI cortical lesion endophenotypes. Based on Nissl staining, we also grouped the ddPCR cohort to the different endophenotypes, and combined them with the MRI animals. Finally, we measured plasma miR-124 level in the combined cohort with reverse transcriptase quantitative PCR (RT-qPCR), and checked if animals with the different lesion endophenotypes also varied in their plasma miR-124 content.

Droplet digital PCR indicated elevation in plasma miR-124 level at 2d post-TBI [~39% more miR-124 copies in TBI animals than controls ($p<0.05$)]. MRI revealed 41% of the 22 rats to possess a “cavity forming” (TBI_{CF}) endophenotype, with larger cortical tissue loss, and an inflammatory rim covering 4% of the ipsilateral cortex. Remaining 59% presented a “chronic inflammatory” (TBI_{CI}) endophenotype, with inflammatory rim covering 11% of the ipsilateral

cortex. Plasma miR-124 analysis from the combined cohort revealed TBI_{CF} to have higher plasma miR-124 at 2d in comparison to both controls (FC=2.09, $p<0.01$) and TBI_{CI} (FC=1.66, $p<0.05$). ROC curve distinguished TBI_{CF} from TBI_{CI} with AUC=0.735 ($p<0.05$), and TBI_{CF} from controls with AUC=0.824 ($p<0.01$). TBI_{CI} did not differ from controls.
Our data indicate that plasma miR-124 is an early biomarker for TBI, and that rats developing the "cavity forming" endophenotype at chronic post-TBI time already present higher plasma miR-124 at 2d post-injury, in comparison to the "chronic inflammatory" group.

Disclosures: S. Das gupta: None. **N. Vuokila:** None. **N. Puhakka:** None. **A. Pitkänen:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.20/CC1

Topic: C.09. Brain Injury and Trauma

Support: NIH K08NS073793

NIH DC IDDRC 1U54HD090257

Mansbach Fellowship

Title: Decreased hippocampal N-Acetylaspartate in rodent models of preterm brain injury is due to mitochondrial dysfunction

Authors: *B. M. TALBOT[1], S. LIN[4], N. NIFORATOS-ANDESCAVAGE, 20010[2], P. WANG[4], S. SCAFIDI[5], J. SCAFIDI[3]
[1]Ctr. for Neurosci. Res., [2]Neonatology, [3]Dept of Neurol. and Ctr. for Neurosci. Res., Children's Natl. Med. Ctr., Washington, DC; [4]Mol. Imaging Lab., Howard Univ. Col. of Med., Washington, DC; [5]Anesthesiol. and Critical Care Med., Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: Long-term cognitive dysfunctions occur in half of children born very premature. Imaging studies in adolescents demonstrate structural dysmaturity and consistent decreased N-acetylaspartate (NAA) - a neurometabolite synthesized specifically by neuronal mitochondria. The cause of decreased NAA, and subsequently the impact of changes in NAA, in the injured developing brain is poorly understood. The objective of this study is to characterize cellular and metabolic alterations that result in decreased NAA in the hippocampus in different rodent models of preterm brain injury. C57BL6/J mice were randomly assigned to hypoxic rearing in a hypoxia chamber (10.5% oxygen) or normal rearing conditions from postnatal day (P) 3-11. At one month of life mice were subjected to a battery of hippocampal-dependent behavioral tests to confirm cognition changes consistent with premature infant cognitive dysfunctions. We

performed ^{1}H-magnetic resonance spectroscopy (^{1}H-MRS) in the hippocampus to quantify neurometabolites, including NAA, using LC-Model. We conducted immunohistochemistry on fixed tissue in that region to compare cell density of post-mitotic neurons between groups. We also assessed group differences in mitochondria by comparing mitochondrial copy number to nuclear DNA. We utilized Western blots to analyze protein expression of mitochondrial-specific proteins and NAA synthesis and degradation enzymes. We are currently looking at differences in precursors to NAA using gas chromatography mass spectrometry, and evaluating NAA synthesis rates with 13C-nuclear MR. Hypoxic mice performed significantly worse in all behavioral tests compared to controls. ^{1}H-MRS revealed significantly decreased NAA levels and NAA/choline ratios in hypoxic mice compared to controls. Together, these results confirm the hypoxia model is appropriate for understanding metabolic and cellular changes due to preterm brain injury. There were no differences in neuronal density, expression of mitochondrial-specific proteins, mitochondrial copy number, or expression of NAA synthesis or degradation enzymes between groups. This data strongly suggest that decreased NAA after hypoxic-induced injury results from altered mitochondrial function. We are further verifying our findings in other models of preterm brain injury, such as the moderate systemic inflammation model using interleukin-1-beta injections. Understanding how NAA production after preterm brain injury relates to mitochondria function can help us develop therapies that target the mitochondria and improve their efficiency.

Disclosures: B.M. Talbot: None. **S. Lin:** None. **N. Niforatos-Andescavage:** None. **P. Wang:** None. **S. Scafidi:** None. **J. Scafidi:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.21/CC2

Topic: C.09. Brain Injury and Trauma

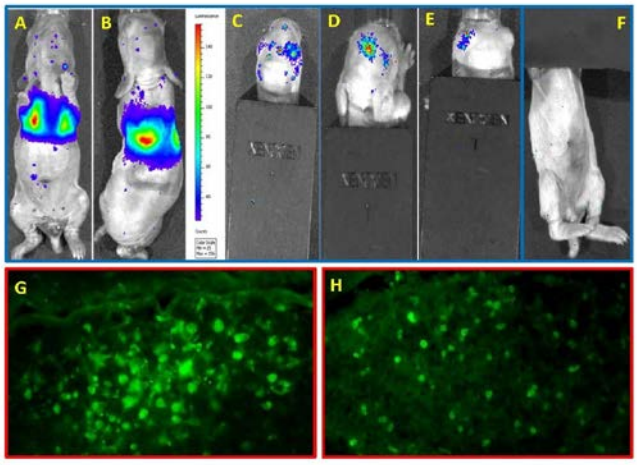
Support: Boston Children's Health Physician's Research Foundation Starter Grant

Title: Improved neurobehavioral outcomes in a rabbit model of germinal matrix-intraventricular hemorrhage following intravenously engrafted human cord blood derived unrestricted somatic stem cells

Authors: *G. VINUKONDA[1,4], Y. LIAO[2], F. HU[3], P. GIRI[5], L. IVANOVA[2], Z. T. MUHAMMAD[3], M. CAIRO[2], E. F. LA GAMMA[6]
[2]Pediatrics-Hematology-Oncology, [3]Pediatrics, [1]New York Med. Col., Valhalla, NY; [4]Pediatrics and Cell bilogy & Anat., New York Med. Col. & BCHP, Valhalla, NY; [5]Pediatrics- Div. of

Newborn Med., Maria Fareri Children's Hospital-New York Med. Col., Valhalla, NY; [6]Dir Regional Neonatal Ctr., Maria Fareri Children's Hosp., Valhalla, NY

Abstract: Germinal matrix-intraventricular hemorrhage (GMH-IVH) is a severe complication of preterm births, which results in acute inflammation and white matter injury leading to cerebral palsy and mental retardation. There is no treatment for IVH or its collateral damage. Stem cells from different sources offer great promise in the treatment of brain disorders. We have isolated novel unrestricted somatic stem cells (USSCs) from human cord blood, with multi-lineage differentiation and regenerative properties. Here we tested if intravenously (i.v.) infused USSC stem cells will: a) survive and migrate in different regions of IVH brain and b) improve neurobehavioral performance in a rabbit model of GMH-IVH. Premature rabbit pups were delivered by Cesarean-section & pups were injected i.p. glycerol (6.5g/kg) to induce hemorrhage. The IVH pups were then infused $1X10^6$ USSC through the jugular vein after confirming IVH by head ultra sound. The USSCs labeled with luciferase reporter gene were monitored for bioluminescence imaging (BLI) on postnatal days 1-3, 7&14. The BLI shows USSCs first appeared in the lung after a few hours of injection & persisted by > 5d in the lung (Fig. A-B). Later the USSCs migrated to the brain starting from day 3 to day 7 (Fig.C-E) to the site of injury. No BLI signals were found in other organs of the body (Fig.F). The migration of USSCs was further confirmed by immunohistochemical staining (IHC) using anti-hNuc antibody specific for USSC stem cells. The immunofluorescence studies of coronal brain section demonstrated the persistence of USSCs at days 7 & 14 forebrain (Fig. G-H). Importantly the motor function and overall neurobehavioral scores were significantly improved in the pups with IVH-USSCs stem cell engrafted intravenously compared with IVH-saline pups at day14 (N=5 in each group ($P<0.05$). These pre-clinical results support the development of a novel stem cell therapeutic strategy for treatment of GMH-IVH human premature infants.



Representative of Bioluminescence imaging & Immunostaining: A-E) BLI-live images for intravenous jugular (i.v.) USSCs infusion demonstrating transient migration via Lung day 3 (A-B) to the brain days 3,5 & 7 (C-E), No USSC in other parts of body (F). G-H) IHC staining using nuclear nuclei specific antibody for USSC stem cells for day7(G) & day17 (H).

Disclosures: G. Vinukonda: None. **Y. Liao:** None. **F. Hu:** None. **P. Giri:** None. **L. Ivanova:** None. **Z.T. Muhammad:** None. **M. Cairo:** None. **E.F. La Gamma:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.22/CC3

Topic: C.09. Brain Injury and Trauma

Support: Center for Neuroscience and Regenerative Medicine

Title: Attenuation of axonal degeneration relative to myelin pathology after mild traumatic brain injury in mice lacking Sarm1

Authors: *C. M. MARION[1,2], K. L. RADOMSKI[2,3], R. C. ARMSTRONG[2,3]
[1]Neurosci. Dept., Uniformed Services Univ., Bethesda, MD; [2]Ctr. for Neurosci. and Regenerative Med., [3]Dept. of Anatomy, Physiol. and Genet., Uniformed Services Univ. of the Hlth. Sci., Bethesda, MD

Abstract: Traumatic brain injury (TBI) results in traumatic axonal injury (TAI) in white matter tracts. Myelin pathology may contribute to deficits experienced after TBI. Myelin pathology may be due to secondary effects along degenerating axons or may occur as primary pathology of white matter injury. The current studies investigate the relationship between axonal degeneration and myelin pathology. Sarm1 is essential for execution of the conserved axon death pathway in injured axons. *Sarm1*-/- mice exhibit suppressed Wallerian degeneration and long-term survival of injured axons. Using a mild TBI (mTBI) model that results in TAI in the corpus callosum, we assessed axon degeneration and myelin pathology in *Sarm1*-/- and wild type mice. The corpus callosum was sampled using electron microscopy 3 days post-mTBI or sham procedure. In wild type mice, TBI increased axon degeneration and produced two forms of myelin pathology - demyelination of intact axons and excessive myelin extending out from axons. In *Sarm1*-/- mice, axon degeneration and demyelination were both suppressed after TBI. However, formation of excessive myelin figures after mTBI was similar in wild type and *Sarm1*-/- mice. We generated myelin reporter mice to label membranes with green fluorescent protein (GFP) to visualize these excessive myelin figures by confocal microscopy. *PLPcreER^{T2};mTmG* mice were given a single low-dose of tamoxifen prior to mTBI/sham procedure, to induce recombination in sparse oligodendrocytes. At 3 days post-injury, aberrant myelin formations were observed along GFP-labeled internodes in mTBI animals as compared to sham controls. These myelin abnormalities may represent dynamic myelin remodeling in a compromised axon-myelin unit. These results indicate that Sarm1 may be an important therapeutic target to interrupt processes that produce axon degeneration and demyelination after mTBI. In addition, mTBI also produces excessive myelin that was not mitigated by suppressing axon degeneration. Funded by the Center for Neuroscience and Regenerative Medicine.

Disclosures: **C.M. Marion:** None. **K.L. Radomski:** None. **R.C. Armstrong:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.23/CC4

Topic: A.10. Development and Evolution

Support: Board of Regents Support Fund LEQSF (2015-18) RDA-14

NIH/NIGMS P30-GM103340

Title: Platelet Activating Factor receptor inhibition improves neurological outcome following pediatric traumatic brain injury

Authors: *J. L. ROSSI, T. TODD, N. G. BAZAN
Dept. of Neurosci., Louisiana State Univ. Neurosci. Ctr. of Excellence, New Orleans, LA

Abstract: Introduction: TBI is known to result in intravascular microthombosis. Children with brain injury had a greater prevalence of disseminated intravascular coagulation compared to non-TBI trauma patients. VWF and ADAMT13 (**a d**isintegrin **a**nd **m**etalloproteinase with a **t**hrombospondin type 1 motif, member **13**) are two proteins known to be involved in the clotting cascade. Platelet activating factor (PAF) is known to contribute to clot formation and this study is to look at the relationship of PAF to VWF and ADAMT13 and how it translates to neurological outcomes following pediatric traumatic brain injury (pTBI). **Methods:** Male PND24 C57bl6 or male platelet activating receptor knockout (PAFrKO) PND24 mice were anesthetized with avertin, mechanically ventilated, physiologically regulated, and subjected to lateral closed-skull injury model with impact depth of 2.00mm, 2.25 mm (bregma level - 0.10mm) or sham. Saline 10ul/g or LAU 901 (a PAF receptor antagonist) - 5mg/kg of LAU901 was injected IP at 12h after impact i.p. Mice were sacrificed at 24h 72h and 7D. Brains were processed with RIPA. Elisa's were then performed on the brain lysates per manufacture's protocols. 2 object novel recognition was performed at 7D before sacrifice to test memory. PAFr KO mice at PND24 7D performed be memory test. **Results: ELISA** *In PND24 saline* injected mice: PAF was statistically elevated at the 72h time point only in the 2mm depths compared to sham and the 2mm was statistically higher than the 2.25mm depth (sham: 6.167 vs. 2mm: 14. 67 vs. 2.25mm: 7.667). VWF was statistically higher in both 2mm and 2.25mm depth compared to sham (Sham: 4.667 vs. 2mm: 11.50 vs. 2.25mm: 12.33) at 72h. ADAMTS13 showed significant elevation in only the 2.25mm compared to sham (Sham: 4.667 vs. 2mm: 9.667 vs. 2.25mm: 14.17) at 72h. *In PND24 LAU* injected mice: PAF was significantly elevated over sham at 24h and 72h in 2.25mm compared to sham (Sham: 4.667 vs 2.25mm: 10.83 and Sham 4.38 vs

2.25mm:11.17) respectively. VWF was only significantly elevated at 72h in the 2.25mm (Sham: 8.167 vs 2.25mm: 14.17) and ADAMTs13 was not elevated at any timepoint or depth. **Behavior** PND24 saline injected sham mice showed preference in the new object vs the old object $p<0.0073$ but the saline TBI mice did not prefer one over the other $p<0.1499$. In the LAU injected PND24 mice the shams and the TBI both showed preference in new vs old objects $p<0.0155$ and $p<0.0036$ respectively. PAFrKO mice also showed preference for the new compared to the old object in both sham $p<0.0001$ and TBI $p<0.0045$. **Conclusion:** Inhibition of PAFr shows changes in proteins related to the clotting pathway are in part dependent upon the PAF receptor and preserves neurological function following pTBI.

Disclosures: J.L. Rossi: None. **T. Todd:** None. **N.G. Bazan:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.24/CC5

Topic: C.09. Brain Injury and Trauma

Support: Canadian Institutes of Health Research (CIHR) MOP 123461

Weston Brain Institute Transformational Grant TR140070

Title: CHIMERA repetitive mild traumatic brain injury induces long-term pathological and behavioral changes in APP/PS1 mice

Authors: *W. CHENG[1], K. M. MARTENS[1], S. STUKAS[1], E. B. BUTTON[1], A. WILKINSON[1], A. BASHIR[1], C. J. BARRON[1], P. A. CRIPTON[2], C. L. WELLINGTON[1]
[1]Dept. of Pathology and Lab. Med., [2]Dept. of Mechanical Engineering, Intl. Collaboration on Repair Discoveries, Univ. of British Columbia, Vancouver, BC, Canada

Abstract: The annual incidence of traumatic brain injury (TBI) is over 2.5 million in the US, with over 3-5 million people living with residual problems. Moderate and severe TBI survivors have high rates of long-term disability and increased risk of neurodegenerative disease. On the other hand, the symptoms of mild TBI (mTBI, the most common form of TBI) usually resolve within weeks of injury. However, some patients may still present symptoms in long-term, which is known as the "Post-concussive syndrome". In addition, repetitive exposure to mTBI has been linked to a neurodegenerative condition called chronic traumatic encephalopathy, which is characterized by tau deposits at sulcal depths and around cerebral vessels, and can exhibit amyloid deposition (50%). Recently, we reported the acute outcomes (6 hr -14 d) of two mTBI (0.5J, 24 hr apart) in the APP/PS1 amyloidogenic mouse model (6-mo or 13-mo), using the

Closed-Head Injury Model of Engineered Rotational Acceleration (CHIMERA). We observed that age at injury, in addition to genetic predisposition to AD, is crucial in determining acute outcomes: TBI induced cognitive deficits and Aβ deposition of APP/PS1 mice in an age-dependent manner; and post-TBI neuroinflammation was exacerbated in 6-mo APP/PS1 but blunted in 13-mo APP/PS1. In the current study, we used the same mTBI paradigm to injure APP/PS1 mice and their wildtype littermates at 6-mo, and assessed the long-term behavioral, histological, and biochemical outcomes of mTBI up to 8-mo post-injury. We found that mice with mTBI had prolonged white matter microgliosis (Iba1) up to 8-mo, and they showed reduced anxiety-like behavior (Elevated plus maze). These two injury outcomes were not different between APP/PS1-TBI and WT-TBI mice. However, long-term fear memory (passive avoidance) was only chronically intensified in APP/PS1-TBI mice. In addition, spatial learning (Barnes maze) was most impaired in APP/PS1-TBI mice, and reversal spatial learning was only impaired in APP/PS1-TBI mice. Neither Aβ nor tau levels in brain homogenates were changed by TBI 8-mo post-injury. These findings suggest that two mild TBI are sufficient to induce long-term neuropathological changes up to 8-mo post-injury without chronic effects on Aβ or tau. Nevertheless, TBI may exacerbate chronic cognitive deficits to a greater extent in mice capable of amyloid deposition.

Disclosures: W. Cheng: None. **K.M. Martens:** None. **S. Stukas:** None. **E.B. Button:** None. **A. Wilkinson:** None. **A. Bashir:** None. **C.J. Barron:** None. **P.A. Cripton:** None. **C.L. Wellington:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.25/CC6

Topic: C.09. Brain Injury and Trauma

Title: A novel mouse model of repeat mild TBI that leads to long-lasting motor deficits and CTE-like brain pathology

Authors: *N. CHO, N. DHILLON, M. ALKASLASI, P. HARO-LOPEZ, O. SHELEST, G. BARMPARAS, E. LEY, G. M. THOMSEN
Cedars-Sinai Med. Ctr., West Hollywood, CA

Abstract: Objectives: Repetitive mild traumatic brain injury (mTBI) has been a suggested risk factor for various neurodegenerative diseases including chronic traumatic encephalopathy (CTE), amyotrophic lateral sclerosis (ALS), and Alzheimer's disease (AD). The use of rodent models to study recurrent mild TBI can provide mechanistic insights into the long-term consequences and help establish a connection between this type of injury and the development of

neurodegenerative disease. We recently developed a novel model of repeat TBI in rats that results in motor deficits sustained out to at least 25 weeks as well as significant brain atrophy and tauopathy consistent with the TBI-linked neurological disease CTE. These sustained deficits and pathology have not been observed in other rodent TBI models that deliver one-time, or repeat, *unilateral* insults. Here, we describe a novel mouse model whereby we delivered a once-weekly, bilateral, closed-skull, controlled cortical impact (CCI) injury over five weeks to reproduce the behavioral and pathological phenotype observed in our rat model, and to recapitulate the human CTE condition.
Methods: We tested various bilateral, closed-skull, CCI injury paradigms, administered once per week for five weeks to C57BL/6J (WT) mice, by varying tip diameter, impact depth and impact velocity. Rotorod testing was performed to assess injury-induced impairments at various time points following injury. Tissue was collected and brain atrophy, as measured by cortical and corpus callosum thinning and tau pathology (AT8) was assessed.
Results: One paradigm tested lead to sustained significant rotorod deficits that persisted for at least 11 weeks post-first injury. Brain tissue collected from a subset of mice euthanized at this time point showed significant brain atrophy (cortical and corpus callosum thinning) and elevated tau pathology that was most extreme in the corpus callosum.
Conclusions: Although motor function deficits are not a prominent feature of acute mild TBI, difficulties in motor coordination are commonly associated with CTE, suggesting that repetitive mild TBI has long-term detrimental effects on the motor system. Here we have provided a unique mouse model of repeat mild TBI that leads to long-term motor impairment with associated CTE-like brain pathology. As we have previously described a similar rat model, the ability to now implement this injury paradigm in transgenic mice will be useful to study repeat mild TBI as a risk factor for a large number of neurodegenerative diseases that are currently modeled in mice but not in rats.

Disclosures: **N. Cho:** None. **N. Dhillon:** None. **M. Alkaslasi:** None. **P. Haro-Lopez:** None. **O. Shelest:** None. **G. Barmparas:** None. **E. Ley:** None. **G.M. Thomsen:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.26/CC7

Topic: C.09. Brain Injury and Trauma

Title: P2X4 and P2X7 receptor blocker in a rat model of cerebral contusion injury

Authors: *M. KOBAYASHI, Y. FURUKAWA, T. KUMAGAWA, K. SHIJO, N. MORO, M. FUKUSHIMA, T. MAEDA, A. YOSHINO
Neurosurg., Nihon Univ. Sch. of Med., Tokyo, Japan

Abstract: Massive release of gliotransmitter ATP is observed immediately after traumatic brain injury. ATP is known to activate nearby microglia which will initiate inflammatory response. P2X4 and P2X7 receptor are the major receptor on microglia to receive ATP signal in vitro that will activate ramified microglia to amoeboid microglia. In the present study, 5-BDBD a selective P2X4 receptor blocker and AZ11645373 a selective P2X7 receptor blocker were used in a in vivo rat cerebral contusion model to observe the magnitude of inflammatory response. Rat cerebral contusion injury was used as a traumatic brain injury model. Control, 5-BDBD, AZ11645373 or both was directly injected into the contused tissue by implanted osmotic pump. Three days after injury, rat was sacrificed and brain was extracted for analysis. Iba-1 antibody positive and Galectin-3 antibody positive amoeboid microglia increased in the surrounding cortical tissue but not in the contralateral hemisphere. Activated microglia was also seen in the ipsilateral hippocampus. Western blotting of Galectin-3 showed significant increase of activated microglia by injury, but this increase was partially suppressed by 5-BDBD and AZ11645373 administration. Some but not all inflammation related cytokines were suppressed by 5-BDBD and AZ11645373 administration. Blockade of microglial activation by P2X4 and P2X7 receptor blocker have an effect to suppress inflammatory response in an in vivo model of traumatic brain injury.

Disclosures: M. Kobayashi: None. **Y. Furukawa:** None. **T. Kumagawa:** None. **K. Shijo:** None. **N. Moro:** None. **M. Fukushima:** None. **T. Maeda:** None. **A. Yoshino:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.27/CC8

Topic: C.09. Brain Injury and Trauma

Title: Function study of knockout brain-specific mitochondrial apoptosis-inducing factor gene (AIF2) in normal and pathological state

Authors: *J. I. RODRIGUEZ[1], Y. ZHANG[1], T. LI[1], Y. SUN[1], C. XIE[1], K. BLOMGREN[2], C. ZHU[1]
[1]Ctr. for Brain Repair and Rehabil., Inst. of Neurosci. and Physiology, Sahlgren, Goeteborg, Sweden; [2]Karolinska Inst., Stockholm, Sweden

Abstract: Perinatal complications such as asphyxia and preterm birth can cause perinatal brain injuries that are often associated with subsequent neurological deficits, such as cerebral palsy. Mitochondria-mediated apoptosis-inducing factor (AIF) dependent neuronal cell death is the central process in perinatal brain injury.
Without an apoptotic stimulus, AIF has a vital role in oxidative phosphorylation and detoxification of reactive oxygen species. However, under apoptotic stimulus, AIF translocate to the nucleus and induce DNA fragmentation and chromatin condensation.
Recently, it has been discovered a brain-specific AIF isoform (AIF2). Numerous functional studies have been performed on AIF1, the most abundant and ubiquitous AIF isoform, whereas, AIF2 has not been further characterized. AIF1 and AIF2 only differ in a short stretch of their amino acid sequence in the N-terminal region that is removed from the mature protein as it translocates to the nucleus. The aim of this study is to assess the functional and regulatory profiling of AIF2 in normal state and after hypoxia-ischemia (HI) insult. For this purpose, we generated AIF2-flox mice and produced AIF2 flox/flox.pgk-cre knockout (KO) mice.
KO AIF2 did not affect the expression of proteins such us Cytc, mia40, OPA1 or SOD2 in neonatal brain mice (9-days old) in physiological state. However, the relative expression of AIF1 in AIF2 KO mice is higher in adult (60-days old) mice, compare to wilt-type litter-mates. KO AIF2 did not affect the fertility of female mice, either caspase-3 activity or the survival and proliferation of neural progenitors in P9 and P60 mice brain. However, KO AIF2 increased brain injury in HI neonatal mice, as showed via immunohistochemistry staining for markers of gray matter (MAP2) at 72 h after HI.
All in all, these data suggest that there is a compensation in the expression level of AIF1 when AIF2 is KO, and this change in the AIF1/AIF2 ratio would lead to an increased brain injury after HI insult.
In summary, we believe that an in-depth understanding of this process will lead to the development of effective treatments during the early stages of perinatal brain injury.

Disclosures: **J.I. Rodriguez:** None. **Y. Zhang:** None. **T. Li:** None. **Y. Sun:** None. **C. Xie:** None. **K. Blomgren:** None. **C. Zhu:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.28/CC9

Topic: C.09. Brain Injury and Trauma

Support: NIH 1-F32NS090748

NIH-NS40125

NIH-NS060672

VAI01RX001127

The Pittsburgh Foundation

Title: Heterozygous cysteine-string protein alpha knock-out mice exhibit reduced SNARE protein abundance following traumatic brain injury

Authors: *S. W. CARLSON, C. DIXON
Neurosurg. and VA Pittsburgh Healthcare Syst., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Traumatic brain injury (TBI) disrupts cognitive function up to 1 year post-injury as reported by patients and observed in preclinical models. Published studies implicate deficits in neurotransmission as contributing to neurobehavioral dysfunction after TBI, but additional work is needed to understand the mechanisms underlying this pathological response. In the synapse, formation of the soluble N-ethylmaleimide-sensitive factor attachment protein receptor (SNARE) complex is an important step facilitating vesicular docking and fusion for neurotransmitter release into the synaptic cleft. Cysteine string protein alpha (CSPα) is a critical chaperone that promotes formation of the SNARE complex. We previously showed CSPα abundance is reduced in the days following TBI. To test the effect of reduced CSPα on neurobehavioral function, we utilized CSPα heterozygous (CSPα HET) mice previously shown to have reduced CSPα protein. Naïve male CSPα HET and wild-type (WT) mice subjected to motor and cognitive testing, performed similarly in the beam balance task. CSPα HET mice exhibited an increase in Morris Water maze spatial acquisition latency on day 3 of testing, compared to WT mice (interaction effect, post-hoc $p<0.05$). We hypothesized CSPα HET mice would exhibit reduced SNARE complex formation and exacerbated neurobehavioral dysfunction after TBI, as compared to WT mice. To test this, CSPα HET and WT mice were subjected to controlled cortical impact (CCI) injury (1.8 mm, 6 m/s, 150 msec dwell) or sham control surgery. Assessment of beam balance performance revealed no differences between groups on days 1-5 post-injury. Assessment of spatial acquisition on days 9-13 post-injury revealed a significant injury effect ($p<0.05$), independent of genotype, compared to sham injury. Evaluation of spatial memory and visible platform performance at 14 days post-injury revealed no difference between groups. Immunoblotting revealed a significant reduction in CSPα abundance in the hippocampus of sham and CCI-injured CSPα HET mice compared to sham and CCI-injured WT mice ($p<0.01$) at 2 weeks post-injury. Assessment of SNARE complex syntaxin-1-immunoreactivity revealed sham and CCI-injured CSPα HET mice had reduced complex formation ($p<0.05$), compared to sham WT mice, at 2 weeks post-injury. While these findings show limited exacerbation of CCI-associated neurobehavioral deficits in CSPα HET mice, future work will investigate compensatory mechanisms capable of promoting SNARE complex formation in the context of reduced CSPα abundance. These data provide novel findings for the effect of TBI on CSPα HET mice and synaptic SNARE protein abundance post-injury.

Disclosures: S.W. Carlson: None. **C. Dixon:** None.

Poster

393. Animal Models of Brain Injury: Molecular Mechanisms, Biomarkers, and Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 393.29/CC10

Topic: C.09. Brain Injury and Trauma

Support: CNRM 307513-10.01-60855

Title: The effect of the angiotensin receptor blocker candesartan after repeated unilateral closed head injury in mice

Authors: M. RUSNAK[1], *A. J. SYMES[2]
[1]USUHS-Henry M. Jackson Fndn., Bethesda, MD; [2]Pharmacol., USUHS, Bethesda, MD

Abstract: The brain renin-angiotensin system (RAS) is an important mediator of inflammation and oxidative stress after CNS injury. Our laboratory and others have shown that treatment with FDA approved angiotensin receptor blockers (ARBs) are effective in ameliorating inflammation and glial reactivity and improving morphological and cognitive recovery after controlled cortical impact injury in mice. As different models of traumatic brain injury (TBI) have distinct neuropathologic features, we wanted to determine whether the ARBs could be effective in a repetitive closed head injury model (rCHI).
In the present study we investigated the effect of the ARB candesartan on behavioral and histopathological outcome after repeated closed head injury. Mice were injured with an electromagnetic impactor on the skin four times, at 24-hour intervals. rCHI produced strong activation of astrocytes and microglia as well as significant loss of neurons in the cerebral cortex in the impacted area compared to the contralateral side 5 days after the last injury. Significant, but less pronounced astrocyte and microglial activation was also observed in the ipsilateral dorsal striatum and corpus callosum. Treatment with candesartan (0.5mg/kg daily) reduced ipsilateral neuronal loss at 5 days post injury but had no effect on astrocyte or microglial activation.
The brain RAS is also involved in the response to stress partially through activation of angiotensin II receptor I (AT1R) in the hypothalamic paraventricular nucleus (PVN). AT1R is co-expressed with corticotropin-releasing hormone (CRH) in the majority of parvocellular neurons of the PVN. We therefore wanted to determine whether candesartan altered the response to restraint stress in rCHI injured mice. rCHI injured mice were subjected to 20 min restraint stress 5 days after the last injury, and sacrificed 2 hours after initiation of the stress. Candesartan treatment reduced CRH mRNA expression in the PVN after 20 min restraint stress compared to vehicle treated mice, but neither the stress nor the candesartan treatment altered AT1R mRNA expression. In behavioral experiments rCHI injured mice showed hyperactivity as compared to

sham animals in open field test. Paradoxically, in the zero maze assay, the injured mice spent more time in the open zones compared to sham animals suggesting a decreased inhibition in an aversive environment, perhaps due to the increased mobility. Candesartan treatment reduced this effect. Thus, in addition to its positive effects improving recovery after controlled cortical impact model, the ARB candesartan has potential benefit in treating closed head injury.

Disclosures: M. Rusnak: None. **A.J. Symes:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.01/CC11

Topic: C.09. Brain Injury and Trauma

Support: NIH Grant ROI NS082432

Dana Foundation David Mahoney Neuroimaging Grant

Title: Soccer heading-related memory deficit in players with the apolipoprotein ε4 allele

Authors: *L. E. HUNTER[1], *L. E. HUNTER[1], M. L. LIPTON[2]
[1]Albert Einstein Col. of Med., Bronx, NY; [2]Dept Radiology, Albert Einstein Col. Med., Bronx, NY

Abstract: Aim: We aimed to determine the extent to which presence of the Apolipoprotein ε4 allele modifies the association between 12-month total number of headers and verbal learning and memory.
Methods: We examined 248 soccer players enrolled the Einstein Soccer Study, a longitudinal study of amateur soccer players in the New York City area. At each visit, players underwent verbal learning and verbal memory testing using Cogstate™ (International Shopping List (ISL) and International Shopping List Recall (ISRL) tasks), respectively. Players also completed Head-Count, a validated, web-based questionnaire that ascertained the total number of headers in the past year. Repeated measures multivariable linear regressions were employed to test for effect modification of ε4 allele status on the association between 12 month total heads and verbal learning and verbal memory.
Results: 24.6 % of the players examined were carriers of at least one copy of the ε4 allele. We found a significant effect modification ($p= 0.007$) of ε4 allele status on the relationship of total headers in the past year and verbal memory (by number of items (1-12) correctly recalled at 20 minutes). Compared to ε4- players with fewest headers of (lowest quartile; 0-130 headers), ε4+ players with most headers (highest quartile; 1044-22838 headers) recalled fewer items = -.99; $p <0.001$). There was a borderline significant effect modification ($p= 0.06$) of ε4 allele status on

the association of heading and immediate recall (i.e., verbal learning).
Conclusions: Soccer players that carry an ε4 allele are more vulnerable than players without the ε4 allele to adverse effects of high levels of heading on memory. These findings suggest that ε4+ players might be targeted in efforts to minimize excessive heading to mitigate potential for adverse impact on memory function.

Disclosures: L.E. Hunter: None. **M.L. Lipton:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.02/CC12

Topic: C.09. Brain Injury and Trauma

Support: James S. McDonnell Foundation grant for the Attention Dynamics Consortium in Traumatic Brain Injury (ADC-TBI)

NIH Grant NS095741

NIH Grant HD083614

NIH Grant DA038834

National MS Society Grant RG 4463A18

Mallinckrodt Institute of Radiology Facilities Pilot Study Fund MIR 14-021

Title: Gradient echo plural contrast imaging detects mtbi related brain tissue damage in areas without evident anatomical changes on clinical mri

Authors: S. V. ASTAFIEV[1], J. WEN[2], D. L. BRODY[3], A. H. CROSS[3], A. P. ANOKHIN[1], K. L. ZINN[3], M. CORBETTA[3], *D. A. YABLONSKIY[4]
[1]Psychiatry, [2]Radiology, [3]Neurol., Washington Univ. in St. Louis, St. Louis, MO; [4]Mallinckrodt Inst. Radiol, Saint Louis, MO

Abstract: Background. It is widely accepted that symptoms in chronic mild traumatic brain injury (mTBI) are caused by injury to the white matter (WM), but the role of grey matter (GM) damage in TBI-related symptomatology is less clear. While WM damage can be detected by DTI, there is currently no accepted technique to detect mTBI-related GM damage. In this study we demonstrate that an advanced implementation of quantitative MRI-based high-resolution 3D Gradient Echo Plural Contrast Imaging (GEPCI) technique(1) sensitive to cellular loss(2,3) allows mTBI-related evaluation of GM tissue abnormalities complementary to DTI.
Methods. We tested 26 healthy controls and 14 mTBI patients with isolated mTBI. GEPCI data

were reconstructed using previously developed algorithms that allow separation of cellular-specific part of relaxation rate constant (R2t*) from global R2* affected by BOLD effect and background gradients. As demonstrated previously, both cell loss in GM and demyelination of WM lead to reduced R2t* values that can be detected by GEPCI.
Results. Single subject voxel-wise analysis (comparing each mTBI patient to 26 control subjects by using a one-sample t-test and selecting voxels with significantly ($p<0.05$) lower R2t* values in mTBI patients) revealed distributed GM abnormalities, not detectable on standard MRI images (T1w, T2w, FLAIR and SWI), in multiple GM regions, especially frontal and temporal, that are frequently damaged after mTBI. We identified 243 clusters of voxels with low R2t* (cluster size: $\geq 27mm^3$, average $634mm^3$) that showed spatial overlap in ≥2 mTBI patients.
To test for random noise, we conducted a similar voxel-wise analysis for control subjects and did not identify any such clusters. Abnormal tissue with the highest degree of overlap across mTBI patients (>45% of overlap) was located in cerebellum, superior temporal gyrus, insula, inferior occipital gyrus, precuneus and inferior temporal gyrus.
Conclusion. We demonstrated that the GEPCI-derived tissue specific R2t* metric is sensitive to GM tissue damage in subjects with mTBI. The ability of GEPCI imaging to detect compromised GM cellular integrity makes it an important diagnostic tool that can complement the information about WM damage provided by the DTI.
1. Luo J, Jagadeesan BD, Cross AH, Yablonskiy DA. NeuroImage 2012;60(2):1073-1082.
2. Ulrich X, Yablonskiy DA. Magn Reson Med 2016;75(2):606-615.
3. Zhao Y, Wen J, Cross AH, Yablonskiy DA. NeuroImage 2016;133:417-429.

Disclosures: S.V. Astafiev: None. **J. Wen:** None. **D.L. Brody:** None. **A.H. Cross:** None. **A.P. Anokhin:** None. **K.L. Zinn:** None. **M. Corbetta:** None. **D.A. Yablonskiy:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.03/CC13

Topic: C.09. Brain Injury and Trauma

Support: National Institute on Disability, Independent Living, and Rehabilitation Research (NIDILRR)

Title: Multimodal tasks can cause more difficulty compared to simple tasks in concussed athletes indicating a dysfunction of the sensorimotor integration mechanisms

Authors: A. B. BIALUNSKA[1], *A. P. SALVATORE[2]
[1]Rehabil. Sci., Univ. of Texas At El Paso, El Paso, TX; [2]U. of Texas At El Paso, El Paso, TX

Abstract: Clinical characteristics of concussion (mild traumatic brain injury, mTBI), include neurological signs and symptoms, as well as cognitive dysfunction which are linked to microstructural injury to neural tissue after biomechanical force to the brain. Such an injury leads to axonal dysfunctions, white matter damage, impaired neurotransmission and potential for neural disconnection (Giza & Hovda, 2014). Cognitive functioning is a complex phenomenon requiring coherent communication across distributed networks of cortical and sub-cortical brain regions. The fine-scale temporal coordination of these regions is critically dependent on white matter tracts that allow effective transmission of information across the brain. Therefore, perturbations following mTBI result in cognitive deficits, but in our opinion particularly in the impairment of multisensory integration. Neurocognitive post-concussion tests were used for assessing verbal and visual memory, attention, and motor processing. However the sensitivity of an instrument for assessing a concussion is a challenge. Sport-related concussion (SRC) is purported to have a relatively short recovery time, 7 to 14 days after injury (DAI) based on an assessment using the ImPACT test. This tool consists of simple, yet not easy tasks. To test whether more complex task which measure multisensory (auditory-visual-verbal-motor) processing could lead to a different evaluation of concussed athletes, we analyzed how the results obtained from two different tests, the ImPACT and Computerized-Revised Token Test (C-RTT), a test of auditory comprehension, change over time following SRC. Using case study method we found that patients showed cognitive dysfunctions both on the ImPACT and C-RTT. Performance was assessed on these instruments in repeated sessions over time. Performance showed a gradual improvement over time. However cognitive performance on ImPACT measures of efficiency and response time were, in some cases, faster than those variables assessed by the C-RTT. The C-RTT measures the accuracy of following spoken sentences and the timing of moving visual objects on a computer screen in response to the spoken sentence. Reaction time performance on the C-RTT was more disrupted and improved much slower than on the ImPACT, about 43 - 60 DAI. Our findings indicate that the more complex task requirements of the C-RTT which required integration of information from different modalities and domains produced poorer performances across time compared to the ImPACT. We hypothesize that the cognitive dysfunction characteristic of SRC is a result of general purpose mechanism (e.g. timing) rather than domain specific.

Disclosures: **A.B. Bialunska:** A. Employment/Salary (full or part-time):; University of Texas at El Paso. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; National Institute on Disability, Independent Living, and Rehabilitation Research (NIDILRR). **A.P. Salvatore:** A. Employment/Salary (full or part-time):; University of Texas at El Paso. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; National Institute on Disability, Independent Living, and Rehabilitation Research (NIDILRR).

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.04/CC14

Topic: C.09. Brain Injury and Trauma

Support: Center for Chronic Disorders of Aging

Title: Neurocognitive, neuroimaging and biomolecular findings in concussed adolescents: An investigation of working memory, math fluency, functional near Infrared spectroscopy (fNIRS), and micro-RNA (miRNA)

Authors: N. SIDEMAN[1,2], H. AYAZ[5], C. J. HAMMOND[2,3], A. SARGENT[5], D. M. APPELT[4,2], S. L. ALLEN[2,1], *B. J. BALIN[4,2]
[1]Psychology, [2]Ctr. for Chronic Disorders of Aging, [3]Div. of Res., [4]Bio-Medical Sci., Philadelphia Col. of Osteo. Med., Philadelphia, PA; [5]Sch. of Biomed. Engineering, Sci. & Hlth. Systems, Drexel Univ., Philadelphia, PA

Abstract: OBJECTIVES: Methods used to make concussion diagnoses, namely neurocognitive testing and symptom reporting/observation, can be unreliable and lack sensitivity. For adolescents, an especially susceptible age group, decisions regarding returning to activities typically rely on these methods. The imprecision of these measures can therefore unnecessarily subject children to further injury and prolonged recovery trajectories. While current methods can help inform some decisions, much of the underlying physiological changes attributed to concussion cannot be gleaned from these standard practices. The current study aimed to evaluate underlying physiological changes that occur in concussions in order to better understand and validate potential bio-marker candidates. **METHODS:** Concussed adolescents (n=10) ages 14-17 within 4 weeks of concussion were compared to matched control subjects (n=10). Subjects completed two tasks (N-back, Math Fluency) while wearing the fNIRS. Each task consisted of three conditions (N-back: N-back1, N-back2, and N-back3; Math Fluency: Addition, Subtraction, and Multiplication). Subjects also provided a saliva sample for miRNA analysis. Groups were compared across neurocognitive, neuroimaging, and biomolecular dimensions. **RESULTS**: Subjects did not differ significantly in age, gender, education, handedness, or hours of sleep. Average age of all participants was 16.1 years (SD=1.33) and mean time since injury was 18.3 days (SD= 6.7). Correlational analyses revealed significant correlations within the response time (RT) and accuracy (ACC) measures of each task. There were significant main effects for the math (p&lt01) and n-back conditions (p&lt01) on RT and ACC across groups. No behavioral differences between groups were seen. Pre-frontal cortex (PFC) brain activation was significantly higher for the mTBI group (p=.049) on the math fluency task. The mTBI group also showed stronger right PFC activation (p=.016) compared to the left PFC. Control subjects did

not show this difference. miRNA responsible for neuro-inflammatory gene expression were regulated differently between groups. **CONCLUSIONS:** This study helps to elucidate the fact that neurocognitive findings often do not vary between individuals with mTBI and their healthy peers. Despite these similar neurocognitive findings between groups, fNIRS and miRNA findings reveal significant underlying physiological differences. These findings support the efficaciousness of neuroimaging and biomolecular analyses in the diagnosis and remediation of concussions. Further work will need to be conducted to analyze the sensitivity and specificity of these methods.

Disclosures: **N. Sideman:** None. **H. Ayaz:** None. **C.J. Hammond:** None. **A. Sargent:** None. **D.M. Appelt:** None. **S.L. Allen:** None. **B.J. Balin:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.05/CC15

Topic: C.09. Brain Injury and Trauma

Support: Canadian Institutes of Health Research (CIHR)

Tomlinson Doctoral Fellowship

Title: Exploring the links between traumatic brain injuries and crime: A 24-year longitudinal study of Canadian males

Authors: *G. GUBERMAN[1,3], M.-P. ROBITAILLE[4,5], A. PTITO[1,2], S. HODGINS[4,5]
[1]Neurol. & Neurosurg., [2]Psychology, McGill Univ., Montreal, QC, Canada; [3]Inst. Universitaire en Santé Mentale de Montréal, Montréal, QC, Canada; [4]Inst. Universitaire en Santé Mentale de Montréal, Montreal, QC, Canada; [5]Psychiatry, Univ. of Montreal, Montreal, QC, Canada

Abstract: Traumatic brain injuries (TBIs) are the leading cause of death and disability in children and young adults around the world (WHO, 2006). Although incidence rates vary across studies, TBIs are estimated to affect approximately 0.6% of Canadians and 1-2% of the US population. Several studies conducted in different countries confirm that the prevalence of TBIs is higher among criminal offenders than in age and sex matched general population samples. However, presently, it is not known whether TBIs precede or follow criminal behavior, hence if they predict criminality. Further, robust evidence from longitudinal investigations shows that from childhood onwards, most offenders display a pattern of conduct problems including impulsive and reckless behaviors. We hypothesized that such behaviors would also increase the risk of TBIs. The present study aimed to determine whether TBIs preceded and predicted criminal offending, and whether the well-known childhood predictors of offending also predicted

TBIs. The study examined 724 males followed from age 6 to 30 in Quebec (Canada), using health records from age 6 to 24 and official criminal records from age 12 to 24. Information was also available on two known criminality-related measures: the mother's age at participants' birth and socioeconomic deprivation of the childhood neighborhood. At age 6, 10, and 12, classroom teachers rated participants' conduct problems, inattention, hyperactivity, hurtful and uncaring behaviors. Our results confirmed previous findings showing that proportionately more offenders than non-offenders had suffered at least one TBI. However, similar proportions of offenders committed their first crime before and after their first TBI. In a logistic regression model that excluded participants with TBIs prior to age 10, experiencing a TBI after 10 was predicted by conduct problems at age 6 (OR 1.07, 95% CI 1.02-1.13), hyperactivity at age 10 (OR 1.28, 95% CI 1.10-1.48), and deprived neighborhood (OR 1.70, 95% CI 1.08-2.68). In models predicting criminal offending, after taking account of childhood behaviors, maternal age, and neighborhood deprivation, sustaining a TBI did not predict subsequent offending. Thus, among males, behaviors observed by classroom teachers at age 6 and 10 and deprivation of the childhood neighborhood predicted subsequent TBIs showing that not all boys are at similar risk for a TBI. Future research is needed to develop more precise measures of childhood behaviors as a first step to establishing programs to prevent TBIs.

Disclosures: M. Robitaille: None. **A. Ptito:** None. **S. Hodgins:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.06/CC16

Topic: C.09. Brain Injury and Trauma

Support: Moody Project for Translational TBI Research

Title: Translational studies of tissue and serum biomarkers for distinguishing between focal and diffuse traumatic brain injury subtypes

Authors: *H. A. WEISZ[1], D. BOONE[1], L. CHERIAN[3], C. ROBERTSON[3], H. S. LEVIN[4], H. SPRATT[2], D. DEWITT[1], D. S. PROUGH[5], H. L. HELLMICH[1]
[1]Anesthesiol., [2]Preventative Med. and Community Hlth., Univ. of Texas Med. Br., Galveston, TX; [3]Dept. of Neurosurg., Baylor Col. of Med., Houston, TX; [4]Cognitive Neurosci Lab., Baylor Col. Med., Houston, TX; [5]Anesthesiol., The Univ. of Texas Med. Br., Galveston, TX

Abstract: Conventional patient neuroimaging often fails to provide sensitive and accurate assessment of brain injury severity or classification. Thus, minimally invasive biomarkers can supplement imaging for patients by identifying prognostic genomic markers specific to brain injury type. We hypothesized that two broad types of traumatic brain injury (TBI), focal or

diffuse, can be distinguished with unique signatures of circulating microRNAs (miRNAs). As a first step, computed tomography (CT) imaging distinguished TBI patients with focal or diffuse injury and later was correlated with serum miRNA levels. RNA was isolated from TBI and non-TBI patient serum, reverse-transcribed, pre-amplified and run on PCR arrays containing 84 common biofluid miRNAs. Fold changes were calculated by comparing TBI to non-TBI controls. One-way ANOVA followed by Benjamin-Hochberg and Tukey's test for multiple comparisons was used to assess statistical significance. Preliminary analysis shows several miRNAs are differentially expressed in human serum between control and injury groups, and one differently expressed miRNA distinguishes between focal and diffuse injury. In parallel animal studies, brain tissue and serum miRNA expression were examined after fluid percussion injury (FPI) and controlled cortical impact (CCI), which produce diffuse and focal injuries, respectively. We hypothesized that unique differential regulation of miRNA in various brain regions would be injury-type specific and that these changes are reflected in serum. Rats were anesthetized and subjected to either severe FPI or CCI and naïve controls remained uninjured. Four hours after TBI, blood was collected, serum was isolated, and rats were sacrificed. Fresh frozen brains were prepared for laser capture microdissection of hippocampus, frontal cortex, and nucleus accumbens. Serum and tissue miRNA were isolated, reverse-transcribed, pre-amplified and run on arrays populated with 84 common disease associated miRNAs. Our analysis shows distinct miRNAs differentially expressed in a brain region dependent manner, between focal or diffuse injuries. Ingenuity Pathway Analysis of differentially expressed miRNA suggests their involvement in potential pathophysiologic processes after TBI. Serum miRNA expression revealed significant differences among FPI and CCI comparisons and to naïve controls. Minimally invasive miRNA biomarkers will improve clinical decision making for TBI patients by supplementing conventional neuroimaging.

Disclosures: H.A. Weisz: None. **D. Boone:** None. **L. Cherian:** None. **C. Robertson:** None. **H.S. Levin:** None. **H. Spratt:** None. **D. DeWitt:** None. **D.S. Prough:** None. **H.L. Hellmich:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.07/CC17

Topic: C.09. Brain Injury and Trauma

Support: Rutgers Stress and Motivated Behavior Institute (SMBI)

Title: Reaction time deficits in asymptomatic adolescents during the subacute period following mild traumatic brain injury

Authors: *K. SPIEGLER[1], J. D. HANDY[2], M. M. CRIPPEN[3], S. KOMARAVOLU[3], K. C. H. PANG[1,4], C. A. MAZZOLA[5], R. J. SERVATIUS[1,2,4,6]
[1]Rutgers Grad. Sch. of Biomed. Sci., Newark, NJ; [2]Syracuse VA Med. Ctr., Syracuse, NY; [4]Dept. of Pharmacology, Physiol. & Neurosci., [3]Rutgers New Jersey Med. Sch., Newark, NJ; [5]Div. of Pediatric Neurolog. Surgery, Goryeb Children's Hosp. of Morristown Med. Ctr., Morristown, NJ; [6]Dept. of Psychiatry, SUNY Upstate Med. Univ., Syracuse, NY

Abstract: Adolescents are at high risk for mild traumatic brain injury (mTBI). Thus, practical and accurate diagnostic tools to evaluate concussive symptoms in this population are of great importance. There is considerable controversy regarding the sensitivity of neurocognitive tests to assess dysfunction in the aftermath of mTBI. Therefore, we examined the performance of adolescents who recently experienced mTBI on the commonly used Immediate Post-Concussion Assessment and Cognitive Testing (ImPACT) and the Defense Automated Neurobehavioral Assessment (DANA), a test used by the military for assessing mTBI. Adolescents ages 11-17 years old with a recent mTBI diagnosis (8 males and 7 females, 9-69 days after concussion) were recruited and compared to non-concussed (NC) volunteers in the same age range (11 males, 3 females). Adolescents completed the online version of the ImPACT battery and completed the DANA battery on a handheld device. There were no differences in self-reported symptoms on the Post-Concussion Symptom Scale comparing mTBI and NC adolescents (Wilks $\lambda = .863$, F (4, 24) = 0.96, $p = .449$, partial $\eta^2 = .137$). With regard to ImPACT performance, the multivariate effect of injury group was not significant (Wilks $\lambda = .821$, F (4, 20) = 1.09, $p = .389$, partial $\eta^2 = .179$). In contrast, there was a significant between-groups multivariate effect for the DANA assessment battery (Wilks $\lambda = .531$, F (3, 20) = 5.88, $p = .005$, partial $\eta^2 = .469$). Follow-up univariate ANOVA revealed significantly lower throughput scores for Simple Reaction Time (F (1, 22) = 13.30, $p = .001$, partial $\eta^2 = .377$) and GoNoGo (F (1, 22) = 7.87, $p = .01$, partial $\eta^2 = .263$) tasks in the mTBI group compared to the NC group. These results suggest reaction time deficits after adolescent mTBI that persist despite the resolution of other symptomatology according to self-report and ImPACT measures. The DANA battery has promise as a tool to determine cognitive dysfunction in the subacute period after mTBI in adolescents.

Disclosures: K. Spiegler: None. **J.D. Handy:** None. **M.M. Crippen:** None. **S. Komaravolu:** None. **K.C.H. Pang:** None. **C.A. Mazzola:** None. **R.J. Servatius:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.08/DP07/CC18 (Dynamic Poster)

Topic: C.09. Brain Injury and Trauma

Support: NIH NINDS R01 NS082432

Dana Foundation David Mahoney Neuroimaging Program

Title: Sex divergence of white matter microstructural change associated with soccer heading

Authors: *T. G. RUBIN[1], E. CATENACCIO[2], R. FLEYSHER[1], L. E. HUNTER[1], N. LUBIN[1], M. E. WAGSHUL[1], W. F. STEWART[3], M. KIM[1], R. B. LIPTON[1], M. L. LIPTON[1]
[1]Albert Einstein Col. of Med., Bronx, NY; [2]Johns Hopkins Univ., Baltimore, MD; [3]Sutter Hlth., Walnut Creek, CA

Abstract: Head impacts in soccer are common. Although men may be exposed to a greater number and greater force of impacts, women report more symptoms which take longer to resolve. These findings suggest that women may be more vulnerable to the effects of heading than men. In a small sample of men and women we previously showed that heading is associated with damage to white matter tract integrity as assessed by lower fractional anisotropy (FA) on diffusion tensor imaging MRI (DTI). In that study of 37 amateur athletes, biological sex was treated as a nuisance covariate. The present study explicitly characterizes the role of sex in the association of heading with microstructural changes (FA) in a cohort of 49 male and 49 female soccer players matched for age and prior 12-month heading. Subjects underwent 3.0T DTI imaging. FA was analyzed with a voxelwise linear model to assess where the association of heading with FA was significant for men, for women, and for significant differences between men and women. We found 3 regions with negative association of heading with FA for men (i.e. decreased FA with heading), and 8 regions with negative association of heading with FA for women. The volume of each region was summed to find 2,121mm^3 of white matter exhibiting lower FA associated with more heading in women, but only 408mm^3 in men. These findings provide evidence that, at the level of brain tissue microstructure, women are more sensitive to the effects of heading than men, supporting the sex differences in symptom frequency and persistence.

Disclosures: T.G. Rubin: None. **E. Catenaccio:** None. **R. Fleysher:** None. **L.E. Hunter:** None. **N. Lubin:** None. **M.E. Wagshul:** None. **W.F. Stewart:** None. **M. Kim:** None. **R.B. Lipton:** None. **M.L. Lipton:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.09/CC19

Topic: C.09. Brain Injury and Trauma

Title: Decreased head movement variability after unilateral vestibular correlates with dizziness: A locomotion study on vestibular schwannoma patients

Authors: *O. ZOBEIRI[1], S. KING[2], R. F. LEWIS[3], K. E. CULLEN[4]
[1]Dept. of Physiol., McGill Univ., Montreal, QC, Canada; [2]Jenks Vestibular Physiol. Lab., Massachusetts Eye and Ear Infirmary, Boston, MA; [3]Otolayngology & Neurol., Harvard Med. Sch., Boston, MA; [4]Johns Hopkins Univ., Baltimore, MD

Abstract: A wide range of functions, from basic reflexes to high-level behaviors, depend on the vestibular system. By sensing head motion and then generating the appropriate reflexes, the vestibular system is vital for maintaining balance and stabilizing gaze. In turn, it has been shown that immediately following unilateral vestibular loss, patients experience impaired balance, postural, and gaze control during clinical tests. However, to date, much less is known about the effects of vestibular loss on voluntary behavior. Here, to assess how unilateral vestibular loss alters voluntary behaviour, we analyzed locomotive behavior in a group of patients with a diagnosis of vestibular schwannoma (VS) who had undergone a primary surgical resection of their tumor via suboccipital craniotomy and retrosigmoid approach with sectioning of the vestibular nerve. Head movements were recorded in healthy volunteers, and patients before the surgery, as well as two and six weeks after surgery using an inertial measurement unit (IMU), which records 3-axis linear acceleration and 3-axis angular velocity. Patients and healthy controls were asked to complete the Functional Gait Assessment, and we focused our analysis on short, 15-to-30-second long normal waling task. We then compared the data that are collected from healthy subjects, as well as patients before and after surgery, to determine if and how patients' movements were altered. Our results confirmed that walking trajectories of patients were more asymmetric than those of control subjects, but were comparable that observed for patients before their surgeries. Patients' gait speed was reduced as compared that observed either before surgery or relative to age matched controls. Specifically, the number of steps generated per second decreased as did the magnitude of forward and vertical acceleration. Surprisingly, two and six weeks after vestibular loss, patients demonstrated a marked reduction movement variability across cycles in both the linear and rotational axes of motion following surgery. Interestingly, our results further showed that patients that demonstrated less variability during walking were actually more impaired; patients that perceived themselves as more dizzy (measured via the Dizziness Handicap Inventory (DHI)) actually demonstrated less reduced variability in their gain during walking. Taken together, our results suggest a positive role for variability in sensori-motor compensation and suggest that movement variability achieved during the early stages of vestibular compensation is an important indicator of how well patients ultimately recover.

Disclosures: O. Zobeiri: None. **S. King:** None. **R.F. Lewis:** None. **K.E. Cullen:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.10/CC20

Topic: C.09. Brain Injury and Trauma

Support: NMU internal funding

Title: Near-infrared spectroscopy measures of prefrontal cortex oxygenation following mild traumatic brain injury

Authors: *T. SUSA[1], K. J. KANGAS[3], M. MOORE[4], J. M. CARLSON[2]
[1]Psychology/Neuroscience, [2]Psychology, Northern Michigan Univ., Marquette, MI; [3]Northern Michigan Univ. Psychology, Marquette, MI; [4]Northern Michigan Univ. Hlth. and Human Performance, Marquette, MI

Abstract: Concussions, or mild traumatic brain injuries, are common in many contact sports. Yet, little is known about the neural consequences of a concussion. Research on the effects of concussions has spiked since 2010. This relatively recent interest has led to an improved understanding of mild traumatic brain injury and how it affects behavior. However, only a small proportion of concussion studies, include a neuroimaging component and in particular, very few studies have assessed the effects of mild traumatic brain injury using functional neuroimaging. This study aimed to fill this knowledge gap by examining prefrontal cortex oxygenation levels during a non-task (i.e., resting) state after mild traumatic brain injury using near-infrared spectroscopy (NIRS). Participants were split into two groups: (1) the experimental group consisted of volunteers who recently suffered a concussion and were then cleared to resume play and (2) the control group consisted of volunteers who were matched based on age, gender, and sport. We recorded using a 16 channel array that was placed over the participant's prefrontal cortex during a five minute rest session. Differences in brain activity were observed across the mild traumatic brain injury and control groups. In general, NIRS activity decreased in the mild traumatic brain injury group across the prefrontal cortex. The results are consistent with previous meta-analytic evidence indicating that the prefrontal cortex is particularly susceptible to concussion. This suggests that NIRS could be used as a portable and cost-effective method of measuring the effects of mild traumatic brain injury on prefrontal cortex activity.

Disclosures: T. Susa: None. **K.J. Kangas:** None. **M. Moore:** None. **J.M. Carlson:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.11/CC21

Topic: C.09. Brain Injury and Trauma

Title: An assessment of frontal lobe activity during an attentional bias task following concussion in collegiate athletes: A near-infrared spectroscopy study

Authors: *K. J. KANGAS[1], J. ADAY[2], M. T. MOORE[3], J. M. CARLSON[2]
[2]Psychology, [3]Sch. of Hlth. and Human Performance, [1]Northern Michigan Univ., Marquette, MI

Abstract: Impacts to the head, that are associated with sports related injuries, can result in a mild traumatic brain injury (mTBI) also known as a concussion. Emotional and attentional impairments often result from concussions; yet, these dimensions of mTBI and their relation to prefrontal cortex (PFC) activity are not well understood. The PFC is involved in increased attentional control during threat-related distractors, and the PFC is particularly susceptible to the effects of mTBI. There is limited research comparing attention-related PFC activation between athletes with and without mTBI in tandem with neuroimaging techniques. To fill this knowledge gap, this study utilized a neuroimaging technique that is inexpensive, non-invasive, and portable to measure PFC activity post-concussion. In particular, near-infrared spectroscopy (NIRS) was used to measure activity during the dot-probe task of affective attentional bias. There were three trial types: baseline (two neutral faces), congruent (dot appears behind the fearful face), and incongruent (dot appears behind the neutral face). This task appears to engage the PFC in past studies. We found significant interactions for trial type and group (mTBI vs Control) as well as time and group. Athletes with mTBI were referred to study personnel, and the control group were matched based on age, gender, and sport. In the control group, the dot-probe task elicited an initial event-related decrease in oxygenated hemoglobin (HbO) in the PFC, which then stabilized and returned to baseline. However, for athletes with mTBI, PFC HbO levels did not dip in response to the task and thus displayed task-insensitivity. At the behavioral level, the mTBI (relative to the control) group showed delayed response times in the dot-probe task. Together, these results allow us to better understand concussions across behavioral and neuroimaging techniques—suggesting the PFC shows an insensitivity to affective stimuli in the dot-probe task of attentional bias after a mTBI.

Disclosures: K.J. Kangas: None. **J. Aday:** None. **M.T. Moore:** None. **J.M. Carlson:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.12/CC22

Topic: C.09. Brain Injury and Trauma

Support: Internal University Support

Title: Lingering dynamic balance decrements in mild traumatic brain injured vs controls in collegiate athletes

Authors: *M. T. MOORE[1], J. M. CARLSON[2], K. J. KANGAS[2], J. P. YOUNG[1], J. J. HAMACHEK[1]
[1]Sch. of Hlth. and Human Performance, [2]Psychology, Northern Michigan Univ., Marquette, MI

Abstract: Balance is a complex process involving visual, vestibular and neuromuscular control. The Biodex Balance System SD is a reliable method to measure dynamic balance. Functional near infrared spectrometry (fNIRS) examines hemodynamic activity in the prefrontal cortex during testing. Little research has examined this low cost approach to testing in mTBI compared to controls after the individual has completed a graduated return to play protocol. Lingering effects after the acute phase could put the individual who is returning to full contact sport at risk for re-injury. The purpose of this research is to examine the differences between mTBI and controls dynamic balance and hemodynamic activity measured by fNIRS 14-42 days after injury and full return to play. Eleven controls matched for age, gender and sport with 11 mTBI (age 18-29) completed dual limb support testing on the Biodex Balance System SD. Limits of Stability (LOS) testing at moderate skill level (75%) involved center of gravity control within their base of support. The clinical test of sensory integration and balance tested stability and sway indexes within four conditions (eyes open/closed on firm vs foam surface) for 30 second intervals. An independent t test revealed significant differences (t(20) p=.033) between controls (M=2.104, SD=.395), and mTBI (M=2.695, SD=.395)] in a dynamic sway balance on a foam surface with the eyes closed. These differences appear to be linked to prefrontal cortex fNIRS activity during dynamic balance tasks. Lingering effects of mTBI beyond the acute phase cause individuals to exhibit difficulty with center of gravity shifts during dynamic settings. Once LOS is exceeded a fall, stumble or step will ensue. This suggests lingering decrements in proprioception and sensory pathways.

Disclosures: M.T. Moore: None. **J.M. Carlson:** None. **K.J. Kangas:** None. **J.P. Young:** None. **J.J. Hamachek:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.13/CC23

Topic: C.09. Brain Injury and Trauma

Support: CIHR MOP-102608

Title: The long-term outcomes of concussion: Alterations in emotional status and executive function

Authors: *V. SICARD[1], *V. SICARD[1], J.-C. LORTIE[1], R. D. MOORE[2], D. ELLEMBERG[1]
[1]Kinesiology, Neurodev Lab. - Univ. of Montreal, Montreal, QC, Canada; [2]Arnold Sch. of Publ. Hlth., Univ. of South Carolina, Columbia, SC

Abstract: Concussions may produce long-term alterations in psycho-affective state, mood and cognition. Moreover, studies suggest that concussion may be associated with self-reported symptoms of executive dysfunction. However, few studies used both subjective and objective measures to assess the long-term outcomes of concussion. Thus, the current study sought to determine whether the cognitive alterations would relate to day-to-day problems in emotional status and behavior. Accordingly, 64 university athletes (32 with a history of concussion (HOC); 32 controls) completed the Beck Depression Inventory II (BDI-II), Beck Anxiety Inventory (BAI), Profile of Mood States (POMS), Behavioral Rating Inventory of Executive Functions for Adults (BRIEF-A), and an experimental task-switching paradigm, which is thought to be a measure of executive functioning. Athletes with a HOC were 6+ months from their last injury, and controls were teammates matched on various factors. Athletes with a HOC reported significantly increased depressive symptoms on the BDI-II (ps < .05) and more problems on the Emotional Control and Self-monitor scales of the BRIEF-A relative to controls (ps < .01). Moreover, they significantly differed on total mood disturbance and all sub-categories except for vigor on the POMS (ps < .05). Athletes with a HOC took significantly longer to respond to respond on the task-switching paradigm (ps < .01). Significant variables were not correlated with the age at which they sustained a concussion, the number of concussions, or time since last injury. The current results suggest that beyond acute and sub-acute phase of injury, not only do athletes with a HOC report greater depressive symptoms and total mood disturbance, but they also exhibit longer reaction times compared to their control teammates. Moreover, they suggest that athletes with a HOC may exhibit more problems in their everyday live such as regulating their emotions and monitoring their behaviors. These alterations may carry negative implications for academic and vocational success and overall well-being.

Disclosures: V. Sicard: None. **J. Lortie:** None. **R.D. Moore:** None. **D. Ellemberg:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.14/CC24

Topic: C.09. Brain Injury and Trauma

Support: C-DAC-FP7/ERC no. 341196 to PV

W81XWH-13-1-0005 to MGH

Title: Altered dynamics of the thalamo-cortical system following mild traumatic brain injury: A combined experimental and theoretical study

Authors: *R. ZUCCA[1], X. ARAKAKI[2], S. C. LOW[1], R. T. GOLDWEBER[3], M. G. HARRINGTON[2], P. F. M. J. VERSCHURE[1,4]
[1]SPECS Univ. Pompeu Fabra, Barcelona, Spain; [2]HMRI, Pasadena, CA; [3]Huntington Mem. Hosp., Pasadena, CA; [4]ICREA, Barcelona, Spain

Abstract: Mild traumatic brain injury (mTBI) is one of the most common neurological disorders and one of the most difficult to diagnose. The initial trauma can lead to a cascade of delayed neurodegenerative events, e.g. diffuse axonal injury and/or excitotoxic neuroinflammation, affecting distal brain areas and causing a variety of adverse sensory, motor, cognitive and affective outcomes which can persist for weeks or even months and lead to severe disability due to cumulative damage (Sharp, 2014; Arakaki, 2016). Because of its diffuse nature, mTBI is difficult to diagnose and no clear biomarkers exist. Electrophysiological methods and computational modelling may shed light on the disruptions in large-scale neural networks involved in the functional deficits following mTBI. Indeed, we have recently shown that stroke-related cortical lesions induce pathological alterations to the thalamo-cortical system, or thalamocortical dysrhythmia (TCD). This arises by attenuating the cortical drive onto the thalamus, switching the latter into a low bursting regime, which further propagates to the neocortex through divergent intra-thalamic circuits (van Wijngaarden, 2016). TCD low-frequency dynamics can account for a variety of non-specific symptoms that are dissociated from the lesion site itself. Here we investigate whether such a network mechanism is implied in mTBI. We investigated brain activation profiles in 10 mTBI patients using resting state EEG after 7, 14 and 31 days following injury, as well as a group of age-matched trauma control patients. Our results show a pattern of increased delta activation in the frontal electrodes, consistent with published results (Lianyang, 2015; Huang, 2009), and attenuation in the beta-band power relative to trauma controls in the acute phase, which gradually recovers after four weeks. At the individual level, the spectral power distributions show high heterogeneity that depends on the lesion site and distribution, suggesting that diagnostically relevant EEG patterns could be revealed taking into account those specific individualized features. To explain the origins of these alterations, we developed a detailed spiking model of the thalamo-cortical circuits to identify the cellular and network mechanisms by which different thalamo-cortical pathways are entrained by means of propagating low-frequency oscillations beyond the restricted region of the diffuse mTBI lesions, giving rise to the associated symptoms.

Disclosures: R. Zucca: None. **X. Arakaki:** None. **S.C. Low:** None. **R.T. Goldweber:** None. **M.G. Harrington:** None. **P.F.M.J. Verschure:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.15/CC25

Topic: C.09. Brain Injury and Trauma

Support: CDMRP MR130266 to LTL

Title: Traumatic photalgia changes in the human brainstem: Tensor-based morphometry

Authors: *L. T. LIKOVA, C. W. TYLER
Smith-Kettlewell Eye Res. Inst., San Francisco, CA

Abstract: Introduction. A well-known consequence of mild, or diffuse, traumatic brain injury (dTBI) is the development of debilitating light-induced pain, or 'photalgia'. The mechanism of the sensitization to light, however, remains unresolved. One hypothesis is that the tissue edema (swelling), which is a common feature of dTBI, might sensitize the trigeminal nucleus to signals from ocular mechanisms activated by bright light.
Methods. To assess this hypothesis, we ran high-resolution Magnetic Resonance Imaging of the brainstem of dTBI participants with mild and severe photalgia, without photalgia, and of healthy controls. The configuration of the brainstem was determined for each participant by Tensor-Based Morphometry (TBM) and compared across the participant groups. The TBM was based on a T1-weighted structural sequence acquired with 0.8 x 0.8 x 0.8 mm voxels. The T1 volume was processed with the FIRST tools from FSL, which provided an affine registration to a standard brain metric (the MNI152), and generated a triangular mesh representation of the brainstem. Data were visualized as a rendering of the surface of the control group colored by the magnitude of the average difference tensor at each vertex, for each of the dTBI groups.
Results and Conclusions. The TBM revealed significant deviations in the brainstem morphology of all dTBI groups. In particular, there was a pronounced difference between the patterns of average swelling and shrinkage between the non-photalgic and photalgic dTBI groups. dTBI without photalgia showed bilateral expansion at the pontine/medulla junction. dTBI with photalgia showed a band of mid-pontine shrinkage, more pronounced in severe photalgia, consistent with deterioration of the trigeminal complex. These results suggest that this shrinkage represents the morphological substrate of the photalgic sensitization of the trigeminal pathway.

Disclosures: L.T. Likova: None. **C.W. Tyler:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.16/CC26

Topic: C.09. Brain Injury and Trauma

Support: DoD

CNRM

Title: Neuroimaging of diffuse axonal and vascular injury in chronic traumatic brain injury

Authors: *M. HABER[1], F. AMYOT[2,4], K. FLESHER[3], K. KENNEY[2,4], C. MOORE[4], E. SILVERMAN[1], Y.-Y. CHOU[5], D. PHAM[5], E. M. WASSERMANN[5], R. R. DIAZ-ARRASTIA[1]
[1]Neurol., Univ. of Pennsylvania, Philadelphia, PA; [2]Neurol., [3]Uniformed Services Univ. of the Hlth. Sci., Bethesda, MD; [4]Ctr. for Neurosci. and Regenerative Med., The Henry M. Jackson Fndn. for the Advancement of Military Med., Bethesda, MD; [5]NINDS, NIH, Bethesda, MD

Abstract: Traumatic Brain Injury (TBI) results in diffuse axonal injury (DAI) and diffuse vascular injury (DVI). Both DAI and DVI result from inertial shearing forces, and the two terms are often used interchangeably, the spatial relationships between DAI and DVI have not been carefully studied. Multimodal magnetic resonance imaging (MRI) can help to distinguish these injury mencahisms: diffusion tensor imaging (DTI) provides information about axonal integrity, while arterial spin labeling (ASL) and functional Blood Oxygen Level Dependent imaging (BOLD) with hypercapnia challenge, reflect cerebral blood flow (CBF) and cerebrovascular reactivity (CVR) respectively. Chronic TBI participants (n=27) and age- and education-matched healthy controls (n=15) underwent multimodality MRI. The Freesurfer image analysis suite (MGH, Harvard, MA) was used to segment each MP-RAGE image into regions of interest (ROIs). Mean values of mean diffusivity (MD), fractional anisotropy (FA), CBF, and CVR were extracted for each ROI. Additionally, maps were normalized into a common space (MNI Atlas) and z-score maps were generated based on a pool of healthy controls. Normality of an ROI/voxel was determined based on z-score (abnormal MD: z-score>2.5; abnormal FA, CBF, and CVR: z-score<-2.5). Abnormal ROIs in one modality were not predictive of abnormalities in another modality. Approximately 8-10% of abnormal voxels for CVR and CBF also show an abnormal voxel value for MD, while only 1% of abnormal CVR and CBF voxels show a concomitant abnormal FA value. These data indicate that chronic TBI patients display two distinct endophenotypes: microstructural tissue/axonal injury and vascular injury that are spatially independent.

Disclosures: **M. Haber:** None. **F. Amyot:** None. **K. Flesher:** None. **K. Kenney:** None. **C. Moore:** None. **E. Silverman:** None. **Y. Chou:** None. **D. Pham:** None. **E.M. Wassermann:** None. **R.R. Diaz-Arrastia:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.17/CC27

Topic: C.09. Brain Injury and Trauma

Title: Derivation of a clinical decision support tool for traumatic brain injury

Authors: *J. E. OLSON[1], M. H. YACYSHYN[1], M. L. WHITMILL[2], M. C. MCCARTHY[2]
[1]Dept Emergency Med., [2]Dept Surgery, Wright State Univ. Boonshoft Sch. Med., Dayton, OH

Abstract: Traumatic brain injury (TBI) is commonly seen in emergency departments and represents a significant burden for health care systems, both during the initial hospital visit and during the recovery period. Numerous clinical models which focus on moderate and severe TBI have been developed to predict long-term outcome following TBI. Because some patients with mild TBI proceed to an unfavorable in-hospital outcome, we previously developed a clinical prediction model for in-hospital mortality which encompasses the entire range of TBI severity. Multiple logistic regression analysis was performed on a derivation data set consisting of 23 clinical parameters retrospectively obtained from 6571 TBI patients presenting to a level 1 trauma center. The analysis yielded eight parameters that are significantly associated with mortality; brain edema, brain contusion, heart rate, systolic blood pressure, age, Glasgow Coma Score, Injury Severity Score, and mode of transportation to the hospital. We are now using the results of this analysis to (1) derive a clinical decision support tool to identify patients likely to have a poor in-hospital outcome and (2) predict patient disposition at discharge. The regression model with beta coefficients calculated from the derivation data set has a receiver operating curve (ROC) with the area under the curve (AUC) of 0.93 and sensitivity of 95% with specificity of 72%. To simplify this model for clinical utility we removed four clinical parameters with the least impact on patient outcome. The ROC of the resulting model has an AUC of 0.90 and a specificity of 53% at a sensitivity of 95%. To test the validity of this model for predicting in-hospital mortality, we collected a validation data set of all eight clinical parameters from patients presenting with TBI to the same level 1 trauma center in 2017. In addition we evaluated the model's ability to predict patient disposition from the hospital using a five-point ordinal scale; 1 = Home, 2 = Rehabilitation facility, 3 = Nursing home, 4 = Hospice care, and 5 = Coroner. With this validation data set, the probability of mortality calculated with our model was higher for patients who died in the hospital. In addition, a more favorable patient disposition was associated with a lower calculated probability of in-hospital mortality ($r^2 = 0.457$, $p < 0.0005$). We conclude

that this model can be used as a clinical decision support tool to identify patients likely to progress to a poor in-hospital outcome and may be useful to predict patient disposition from the hospital. This will aid the allocation of hospital resources and alert physician to patients which may need early interventions.

Disclosures: J.E. Olson: None. **M.H. Yacyshyn:** None. **M.L. Whitmill:** None. **M.C. McCarthy:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.18/CC28

Topic: C.09. Brain Injury and Trauma

Support: Grant of Russian Scientific Found N 17-15-01487

Title: BLOOD biomarkers of excitotoxicity in children with traumatic brain injury

Authors: *V. G. PINELIS[1], E. SOROKINA[2], E. ARSENIEVA[2], A. SURIN[2], J. SEMENOVA[3], O. KARASEVA[4], V. REUTOV[5], L. ROSHAL[4]
[1]of neurobiology, Scientific Ctr. For Children s Health, Russian M, Moskva, Russian Federation; [2]Lab. of neurobiology, Natl. Scientific Ctr. for Children's Hlth., Moscow, Russian Federation; [3]neurosurgery, Inst. of Emergency Children' s Surgery and Traumatology, Moscow., Moscow, Russian Federation; [4]Inst. of Emergency Children' s Surgery and Traumatology, Moscow, Russian Federation; [5]Inst. of Higher nervous activity and Neurophysiol., Moscow, Russian Federation

Abstract: Traumatic brain injury (TBI) is often accompanied by hypoxia and inflammation of brain tissue. The cascade of harmful changes in neurons and glial cells followed by an excess concentration of glutamate in synaptic gap, glutamate receptors overstimulation, increased influx to neurons Ca^{2+} and Na^{+}, oxidative stress and the development of autoimmune response may be responsible for a second impact brain injury. The aim of this study was to evaluate the diagnostic and prognostic significance of different brain markers in blood of children with TBI within 24-48 hours after trauma. The severity of TBI was evaluated according to the Glasgow Coma Scale. Based on the clinical examination children following TBI were divided into 2 groups: (1) mild TBI (35 children with concussion; GCS=15); and (2) moderate and severe TBI (25 children; GCS=8-12). TBI markers such as S100b, autoantibodies (aAB) to NMDA and AMPA receptors (NMDARc and AMPARc), 3-nitrotyrosine were measured in blood serum; NMDARc and AMPARc peptides were measured in plasma using ELISA methods. The levels of $NO^{\cdot}$ metabolites were measured with assay kit from Calbiochem. Results: 1) Mild TBI was characterized by high heterogeneity of changes in S100b than might be due to trophic or

adaptation features of some elevation of S100b. The elevated levels of S100b in the first days after TBI in patients from second group were not strictly correlated with severity of TBI and fell to normal levels after 2-3 days; 2) The increased level of aAB to NMDARc observed on the 1st day after TBI in children with concussion. At the same time in patients from second group the level of aAB to NMDARc was decreased; 3) The normal level of NMDARc peptide was detected in all observed children; 4) The increased concentration of AMPARc peptide was revealed only in children with concussion. The level of this peptide in children from the 2nd group was the same as in healthy children. It should be noted that the level of antibodies to AMPARc in all examined children was in the range of normal values. The observed increase in the levels of AMPA peptide and autoantibodies to NMDA receptors in children with concussion should be considered in terms of the risk of future neurological complications. 5) The levels of NO˙ metabolites and 3-nitrotyrosine were significantly increased in all children with TBI especially in patients with severe TBI. Thus, our studies of chosen parameters allowed to form a panel of biomarkers for the diagnosis and prognosis of TBI in children. The revealed changes are also important for clarifying the mechanisms of brain damage in children with TBI. This work was supported with grant of Russian Scientific Found N 17-15-01487.

Disclosures: **V.G. Pinelis:** A. Employment/Salary (full or part-time):; National Scientific Center for Children's Health, Russian Ministry of health. **E. Sorokina:** A. Employment/Salary (full or part-time):; National Scientific Center for Children's Health, Russian Ministry of health. **E. Arsenieva:** A. Employment/Salary (full or part-time):; National Scientific Center for Children's Health, Russian Ministry of health. **A. Surin:** A. Employment/Salary (full or part-time):; National Scientific Center for Children's Health, Russian Ministry of health. **J. Semenova:** A. Employment/Salary (full or part-time):; Institute of Emergency Children' s Surgery and Traumatology. **O. Karaseva:** A. Employment/Salary (full or part-time):; Institute of Emergency Children' s Surgery and Traumatology. **V. Reutov:** A. Employment/Salary (full or part-time):; Institute of Higher nervous activity and Neurophysiology. **L. Roshal:** A. Employment/Salary (full or part-time):; Institute of Emergency Children' s Surgery and Traumatology.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.19/CC29

Topic: C.09. Brain Injury and Trauma

Support: H2020 SME Phase 2 ComAware

Title: Command following assessment and communication with vibro-tactile p300 and motor imagery brain-computer interface tools in complete locked-in and locked-in patients

Authors: *C. GUGER[1], R. SPATARO[2], B. ALLISON[1], A. HEILINGER[1], R. ORTNER[3], W. CHO[3], V. LABELLA[2]
[1]Guger Technologies OG, Graz, Austria; [2]Univ. of Palermo, Palermo, Italy; [3]g.tec medical engineering GmbH, Schiedlberg, Austria

Abstract: Many patients with locked-in syndrome (LIS) or complete locked-in syndrome (CLIS) also need brain-computer interface (BCI) platforms that do not rely on visual stimuli and are easy to use. We investigate command following and communication functions of mindBEAGLE with 9 LIS, 3 CLIS patients and three healthy controls. This tests were done with vibro-tactile stimulation with 2 or 3 stimulators (VT2 and VT3 mode) and with motor imagery (MI) paradigms. In VT2 the stimulators are fixed on the left and right wrist and the participant has the task to count the stimuli on the target hand in order to elicit a P300 response. In VT3 mode an additional stimulator is placed as a distractor on the shoulder and the participant is counting stimuli either on the right or left hand. In motor imagery mode the participant is instructed to imagine left or right hand movement. VT3 and MI also allow the participant to answer yes and no questions. Healthy controls achieved a mean assessment accuracy of 100% in VT2, 93% in VT3, and 73% in MI modes. They were able to communicate with VT3 (86.7%) and MI (83.3%) after 2 training runs. The patients achieved a mean accuracy of 76.6% in VT2, 63.1% in VT3, and 58.2% in MI modes after 1-2 training runs. 9 out of 12 LIS patients could communicate by using the vibro-tactile P300 paradigms (answered on average 8 out of 10 questions correctly) and 3 out of 12 could communicate with the motor imagery paradigm (answered correctly 4,7 out of 5 questions). 2 out of the 3 CLIS patients could use the system to communicate with VT3 (90 and 70% accuracy). The results show that paradigms based on non-visual evoked potentials and motor imagery can be effective for these users. It is also the first study that showed EEG-based BCI communication with CLIS patients and was able to bring 9 out of 12 patients to communicate with higher accuracies than reported before. More importantly this was achieved within less than 15-20 min.

Disclosures: **C. Guger:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Guger Technologies OG. **R. Spataro:** None. **B. Allison:** A. Employment/Salary (full or part-time):; Guger Technologies OG. **A. Heilinger:** A. Employment/Salary (full or part-time):; Guger Technologies OG. **R. Ortner:** A. Employment/Salary (full or part-time):; g.tec medical engineering GmbH. **W. Cho:** A. Employment/Salary (full or part-time):; g.tec medical engineering GmbH. **V. LaBella:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.20/CC30

Topic: C.09. Brain Injury and Trauma

Title: Clinical severity in the acute phase and brainstem volume reduction in the chronic phase in diffuse axonal injury

Authors: *G. FUJIMOTO[1], S. UBUKATA[2], N. OISHI[3], T. ASO[3], G. SUGIHARA[2], T. MURAI[2], K. UEDA[2]
[1]Psychiatry, Kyoto Univ. Grad. Sch. of Med., Kyoto-Shi, Japan; [2]Psychiatry, Kyoto Univ. Grad. Sch. of Med., Kyoto, Japan; [3]Human Brain Res. Center, Kyoto Univ., Kyoto, Japan

Abstract: Diffuse axonal injury (DAI), one of the major forms of traumatic brain injury, is characterized by diffuse white matter disruption caused by shearing forces on the brain. Loss of consciousness is present in the acute phase, and some behavioral disorders or psychiatric symptoms are often apparent in the chronic phase. One of the significant image findings in the chronic phase is atrophy of brainstem and cerebrum, which is thought to reflect acute severity, but there are few reports that focused on the correlation between chronic reduction in volume of brainstem or cerebrum and acute severity. The aim of this study is to investigate the normal volume of each brain part based on healthy subjects' data, and to assess the correlation between the estimated reduction in volume of each part and acute severity in DAI. Twenty patients with DAI and sixty age- and gender-matched healthy controls underwent T1-weighted three-dimensional magnetization-prepared rapid gradient-echo (3D-MPRAGE). 3D-MPRAGE data were processed by Freesurfer to determine individual volumes of interest. First, we performed group comparison, and identified the statistically significant difference in the volume of brainstem and cerebrum between healthy subjects and DAI patients. Second, we performed correlation analysis on healthy subjects' data between each individual's intracranial volume (ICV) and volume of cerebrum, midbrain, pons, and medulla. Every volume of these brain parts above was significantly correlated with ICV, but the more distal the brain part was, the smaller its coefficient of correlation value was. Third we calculated the estimated atrophy rate in each brain part in DAI individuals by comparing the measured values of volume with the estimated preinjury volume which we got based on multiple-regression analysis on healthy subjects' data. The estimated atrophy rate was significantly larger in brainstem than in cerebrum, and the atrophy rate of brainstem showed a significantly positive correlation to an acute severity, the duration of post-traumatic amnesia. These results suggest that chronic atrophy rate of brainstem can be a marker for estimating the acute severity, and that acute severity may be a marker for predicting chronic atrophy of brainstem.

Disclosures: **G. Fujimoto:** None. **S. Ubukata:** None. **N. Oishi:** None. **T. Aso:** None. **G. Sugihara:** None. **T. Murai:** None. **K. Ueda:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.21/CC31

Topic: C.09. Brain Injury and Trauma

Support: NIH 9500-306-270

Title: Alterations in perivascular Aquaporin-4 distribution in chronic traumatic encephalopathy

Authors: *B. R. HUBER[1], V. VENTRANO[2], B. KNAPP[2], J. D. CHERRY[3], V. ALVAREZ[1], T. STEIN[2], A. C. MCKEE[4]
[1]VA Boston Healthcare, Boston, MA; [2]Boston Univ. Med. Sch., Boston, MA; [3]Neurol., Boston Unviersity, Allston, MA; [4]Boston Univ., Boston, MA

Abstract: Chronic traumatic encephalopathy (CTE) is a neurodegenerative disease associated with repetitive exposure to traumatic brain injury (TBI). It is characterized by perivascular hyperphosphorylated tau (ptau) neurofibrillary tangles, and astrocytic ptau deposits at the sulcal depths. The glial lymphatic (glymphatic) system is a waste clearance system that facilitates perivascular fluid flux through aquaporin-4 (Aqp4) channels. Most normal vessels in the brain have perivascular astrocytes with processes that ensheathe vessels with astrocytic end-feet. Recent studies suggest that end-feet regulate fluid flow and are part of the glymphatic system. Animal models have demonstrated that aquaporin-4 redistributes from astrocyte end-feet to cell body after traumatic brain injury (TBI). The perivascular distribution of aggregated tau and involvement of astrocytes in CTE suggests that astrocytes in the glymphatic system may be altered in CTE. To address this hypothesis, we examined the distribution of glial fibrillary acidic protein (GFAP) and Aqp4 in individuals with CTE neuropathology. We found that Aqp4 positive astrocytes were depleted in the perivascular space. The remaining perivascular astrocytes expressed increased GFAP indicative of reactive astrocytosis. Moreover, Aqp4 positive end-feet were depleted around vessels with aggregated tau. These findings demonstrate alteration of the glymphatic system within CTE lesions. We also found an increase in activated perivascular microglia and reactive astrocytes in the perivascular space suggesting an ongoing inflammatory process. These findings demonstrate alterations of the glymphatic system within the perivascular tau lesions considered pathognomonic for CTE.

Disclosures: B.R. Huber: None. **V. Ventrano:** None. **B. Knapp:** None. **J.D. Cherry:** None. **V. Alvarez:** None. **T. Stein:** None. **A.C. McKee:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.22/CC32

Topic: C.09. Brain Injury and Trauma

Title: Concussion and a single season of contact sport participation affect performance on a test of high memory interference

Authors: *M. D. MCCRADDEN[1], S. BECKER[2], P. I. ROSEBUSH[3], M. F. MAZUREK[4]
[1]Neurol., [2]Psychology Neurosci. and Behaviour, [3]Dept Psych & Behav Neuro, McMaster Univ., Hamilton, ON, Canada; [4]Dept Med. & Neurosci, McMaster Univ. Med. Ctr., Hamilton, ON, Canada

Abstract: Introduction:
Sport-related concussion is one of the most impactful injuries sustained by youth and varsity athletes. Recent attention has also been paid to the effects of contact sport participation in the absence of concussion, and the potential cognitive effects of repeated 'subconcussive' head injuries.
The hippocampus is known to be vulnerable to head injury. A test involving a high memory interference component, known as the mnemonic similarity test (MST), may be able to detect deficits of hippocampal neurogenesis. The goal of this project was to utilize the MST to investigate cognitive changes in: 1) concussed athletes and 2) contact athletes sustaining subconcussive impacts over a single rugby season.
Methods:
A total of N = 160 McMaster athletes participated between September 2014-January 2016. During the preseason, all individuals completed sport psychology questionnaires and a semi-structured psychiatric interview. We then administered the MST, which tests recognition of images that are new, old, or very similar previously presented images. Two subgroups were studied: 1) *Concussion*: 11 athletes in the original cohort supplemented by 6 others referred to our neurology clinic were matched using a 1:2 ratio with athlete controls and were tested while symptomatic (~2-4 weeks postinjury) and again upon recovery; 2) *Contact athletes*: in addition to the preseason assessment, 13 rugby players also completed midseason and postseason testing.
Results:
1) In the *concussion* subgroup, we found impaired identification of similar stimuli during the symptomatic phase, which had significantly improved upon full concussion recovery. The identification of old stimuli was also mildly impaired while symptomatic, and also improved. Using a regression model we
found that concussion was significantly predictive of the degree of impairment in the identification of similar stimuli.

2) In the *contact athlete* subgroup, we found a significant impairment of their ability to identify similar stimuli during the midseason, but in the postseason this resolved and surpassed preseason performance.
Significance: This study is the first to utilize the MST in concussion and contact sport samples. We found that the ability to discriminate between highly similar stimuli is impaired following concussion, and with contact sport participation - in both subgroups, test performance improved after a reprieve from sport participation. This study contributes to the growing body of evidence indicating subconcussive head impacts may affect cognition in the absence of an overt concussion. These results may reflect a head- injury-induced impairment of hippocampal neurogenesis.

Disclosures: M.D. McCradden: None. **S. Becker:** None. **P.I. Rosebush:** None. **M.F. Mazurek:** None.

Poster

394. Traumatic Brain Injury: Human Studies II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 394.23/CC33

Topic: C.09. Brain Injury and Trauma

Support: CDMRP 13129004

PT074437

CNRM Grant 300606-7.01-60855

NARSAD, Brain Behavior Research Fund Grant 18317

Title: Predicting military-related mild traumatic brain injury disease - a multimodal neuroimaging study with machine learning

Authors: P.-H. YEH, C. G. KOAY, J. GRANER, *S. RAJAMONI NADAR, W. LIU, T. OAKES, G. RIEDY, G. BONAVIA, J. OLLINGER
Natl. Intrepid Ctr. of Excellence, Walter Reed Natl. Military Med. Ctr., Bethesda, MD

Abstract: Purpose:
Machine learning methods using neuroimaging data for the diagnosis of neurodegenerative and psychiatric disorders have been the subject of extensive research in recent years. The goal of this study is to demonstrate the feasibility of applying machine learning techniques that can be useful in diagnosing service members with mild traumatic brain injury (mTBI).
Materials and Methods:
Participants included 130 male active service members diagnosed with mTBI (age 34.7±7.8

years old) and 53 non-TBI male controls (age 31.9±8.3 years old) who underwent neuroimaging exams on a 3T MRI scanner. Several metrics derived from neuroimaging modalities, including diffusion tensor imaging (DTI), dynamic susceptibility contrast perfusion MRI and resting state fMRI, were used as predictors. After regressing out age effect, the accuracy of identifying mTBI patients using multimodal neuroimaging metrics was predicted using several machine learning techniques, including linear logistic regression, random forest, Gaussian Naïve Bayesian classifier, support vector machine (SVM) followed by feature transform and feature selection (k highest scoring features), and cross validation. Machine learning algorithms from Scikit-Learn package (http://scikit-learn.org) were used for post-processing classification.

Results:

The accuracy of identifying mTBI patients varied from 67% to 82% with relative consistency of accuracy of around 73% ± 4 % using the method of SVM followed by feature transform and feature selection, and cross validation; however, the accuracy of identifying mTBI using the metrics derived from a single neuroimaging modality varied among different modalities. The important features in identifying mTBI include clusters of DTI metrics and relative cerebral blood flow over the internal / external capsule, thalamus, corpus callosum, cerebral peduncles, cingulum bundle, corona radiata; and the clusters of functional networks, including the posterior / anterior default networks, bilateral frontoparietal networks, salience network, and the central executive network.

Discussion and conclusions:

These preliminary results suggest machine learning techniques might be useful in identifying mTBI with acceptable accuracy. Our results support disrupted neurocircuitry in mTBI, particularly involving the cortico-thalamo-striatal networks interconnecting frontal cortex, parietal cortex and thalamostriatal system. Future work, including adding additional neuroimaging metrics, neurobehavioral data, and using more advanced techniques such as deep learning techniques with a larger sample size for cross validation has been under way.

Disclosures: P. Yeh: None. **C.G. Koay:** None. **J. Graner:** None. **S. Rajamoni Nadar:** None. **W. Liu:** None. **T. Oakes:** None. **G. Riedy:** None. **G. Bonavia:** None. **J. Ollinger:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.01/DD1

Topic: C.09. Brain Injury and Trauma

Title: Minocycline plus N-acetylcysteine targets synaptic plasticity and restores cognition when first dosed three days after closed head injury

Authors: *K. WHITNEY, E. NIKULINA, L. BUITRAGO-SOTO, P. J. BERGOLD
Dept. of Physiol. and Pharmacol., State Univ. of New York Downstate Med. Ctr., Brooklyn, NY

Abstract: Traumatic brain injury (TBI) produces long-lasting cognitive and behavioral deficits. Patients often wait days to seek treatment, only when their symptoms do not abate. Therefore, an effective drug to treat TBI must retain potency when dosed days after an injury. We tested the potency of minocycline (MINO) plus N-acetylcysteine (NAC) when first dosed 72 hours in the mouse closed head injury (CHI) model. CHI produces many of the same outcomes as TBI, including heterogeneous severity, cognitive and behavioral deficits, and gray and white matter damage both ipsilateral and contralateral to the impact site. The hippocampus is particularly susceptible to both TBI and CHI. CHI damages the ipsilateral and contralateral hippocampi. Both hippocampi display neuronal loss, decreased expression of synaptic markers, decreased dendritic density and altered spine morphology. Fourteen days after CHI, the contralateral hippocampus has decreased expression of neuronal marker NeuN, the pre-synaptic marker synaptophysin, and post-synaptic markers PKMζ and MAP2 in areas CA1 and CA3. Injured mice are impaired in acquisition of Barnes maze, a spatial memory task that requires one intact hippocampus. LTP is impaired in Schaffer collateral-CA1 synapses in slices isolated from both the ipsilateral and contralateral hippocampus. MINO plus NAC treatment beginning at 72 hours after CHI improved performance on Barnes maze and prevented neuronal loss in both hemispheres, suggesting the drugs are acting both at and distal to the impact site. Treatment maintained hippocampal dendritic structure, spine density and morphology, and preserved expression of pre- and post-synaptic markers. MINO plus NAC also restored Schaffer collateral LTP in the contralateral hippocampus. These data suggest that: (1) CHI induces gray matter injury leading to long-term changes to neuronal structure and synaptic connections; (2) these long-term changes result in impairments in synaptic plasticity and hippocampus-dependent behavior; and (3) MINO plus NAC first dosed 72 hours after injury limits these impairments. Our results identify dendritic morphology and synaptic plasticity as potential targets for drugs that can be dosed at clinically useful windows to improve functional outcome after CHI. Both MINO and NAC are FDA-approved drugs suggesting that this combination could be used to limit cognitive and behavioral deficits after clinical TBI.

Disclosures: K. Whitney: None. **E. Nikulina:** None. **L. Buitrago-Soto:** None. **P.J. Bergold:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.02/DD2

Topic: C.09. Brain Injury and Trauma

Title: Therapeutic benefit of galantamine in hyperoxic brain injury in neonatal mice

Authors: *N. ZAGHLOUL[1,2], N. COHEN[2], K. AYYASOLA[2], V. PAVLOV[3], M. N. AHMED[2]
[1]Pediatrics, Cohen's Children Med. Ctr., Manhasset, NY; [2]Neonatal-Perinatal Med., [3]Ctr. for Biomed. Sci., Feinstein Inst. for Med. research, Manhasset, NY

Abstract: Background: Brain growth and development in premature infants is affected by hyperoxic environments. Hyperoxia leads to excess free radical production with subsequent inflammation, astrogliosis, microgliosis and apoptosis. Galantamine, an acetylcholinesterase inhibitor, has been studied previously for its role in reducing hypoxic brain injury through its anti-inflammatory effects. **Objective:** To explore the role of galantamine in reducing hyperoxic brain injury in neonatal mice. **Design/Methods:** Wild type mouse pups were placed in a hyperoxia chamber (FiO2 95%) for 7 days. Half the group was injected daily with intraperitoneal galantamine (5mg/kg/dose) and the other half was injected with normal saline. After exposure, mice were sacrificed and brain tissue was studied. To study cholinergic neurons and glia, we performed immunflorescent (IF) staining for choline acetyl transferase (ChAT), NeuN, Iba-1, CD68, CNPase and GFAP. Inflammatory markers; IL-1β, IL-6, TNF-α; were measured by Elisa and NF-κB activity (phosphorylated P65) was assessed by western blot. Acetylcholinesterase activity was assayed. Caspase 3 activity, a marker of apoptosis, and reactive oxygen species were assessed fluorometrically. All results were compared to control group housed in room air. **Results:** There was a statistically significant increase in ChAT (IF and WB) and a decrease in acetylcholinesterase activity in the hyperoxia groups treated with galantamine compared to those treated with saline ($P<0.05$). The preservation of ChAT neurons in the galantamine-treated hyperoxic group was associated with a significant decrease in microgliosis, astrogliosis and NF-kB activation. In the galantamine treated hyperoxia group, oligodendrocytes were preserved and therefore myelination. Both ROS and studied inflammatory cytokines; IL-1β, IL-6, and TNF-α; were statistically decreased in the galantamine treated hyperoxia group when compared to the saline treated hyperoxia group ($P<0.05$). **Conclusion(s):** Galantamine has potent anti-inflammatory and antioxidant short-term effects in hyperoxia-induced brain injured neonatal mice. Galantamine attenuates the inflammatory response by decreasing brain cytokine levels, microgliosis, NF-kB activation, and oxidative stress induced by hyperoxia. Galantamine decreases oligodendrocyte loss thus preserving myelination. This data suggests a potential role for galantamine in attenuating hyperoxia-induced brain injury.

Disclosures: N. Zaghloul: None. **N. Cohen:** None. **K. Ayyasola:** None. **V. Pavlov:** None. **M.N. Ahmed:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.03/DD3

Topic: C.09. Brain Injury and Trauma

Support: SUNY Downstate Alumni Association

Title: Altered cognition, anxiety, and neuroinflammation in mouse model of comorbid traumatic brain injury and post traumatic stress disorder

Authors: *K. ST. LAURENT-ARIOT[1], A. FESHARAKI[2], J. MIYAUCHI[3], S.-A. E. TSIRKA[4], P. J. BERGOLD[5]
[1]SUNY Downstate COM, Brooklyn, NY; [2]Yale Univ. Sch. of Med., New Haven, CT; [3]Dept. of Pharmacol., Stony Brook Univ., Stony Brook, NY; [4]Stony Brook Univ. Med. Ctr., Stony Brook, NY; [5]SUNY-Downstate Med. Cen., Brooklyn, NY

Abstract: Comorbid traumatic brain injury (TBI) and post-traumatic stress disorder (PTSD) have been understudied despite increasing evidence that it produces greater psychological and neurological deficits than TBI or PTSD alone. TBI or PTSD produce long-lasting neuroinflammation. A key question is: Does TBI, PTSD or co-morbid TBI and PTSD produce differing levels of neuroinflammation. C57Bl/6mice (28g) received either closed head injury (CHI), a model of TBI; chronic variable stress (CVS), a model of PTSD; CHI followed by CVS (CHI→CVS), or CVS followed by CHI (CVS→CHI), both models of co-morbid TBI/PTSD. CHI is produced by a single strike to the skull over the parietal lobe. CVS is four days of chronic variable stress consisting of cold-water swim, transient restraint, cage tilt, wet cage, and food deprivation. Barnes Maze and active place avoidance assayed cognition and memory. Elevated plus maze evaluated basal anxiety and acoustic startle response assayed fear-potentiated anxiety. Mice in the CHI, CVS, and CHI→CVS groups had similar acquisition of Barnes maze and active place avoidance as the sham-CHI and CVS groups, suggesting no deficits in cognition. However, CVS→CHI mice did not acquire active place avoidance suggesting a deficit in cognition. On elevated plus maze, the CVS group had significantly higher basal anxiety. All groups, except for CHI→CVS, did not differ in the frequency of freezing as intensity increased from 90dB to 105dB. Neuroinflammation was assessed in mice sacrificed 12 days after the onset of CHI or CVS. Expression of glial acidic fibrillary protein (GFAP) assayed astrocyte activation, Iba-1 assessed microglial activation, and the mitochondrial transporter TSPO assessed overall changes in inflammation. All experimental groups increased GFAP expression in the amygdala. In contrast, Iba-1 expression was only significantly higher in the dentate gyrus, CA3 and CA1 regions of the CVS→CHI group. The hippocampus of the CVS→CHI group had significantly higher expression of the M2 marker Arg-1^{hi} as well as increased TSPO expression. These data suggest an increased inflammatory response in the CVS→CHI group. The CVS→CHI group was treated with the TSPO agonist AC5216 to test for a potential anti-inflammatory or anxiolytic role of TSPO. Preliminary data suggests AC5216 lowered anxiety, but had no effect on cognition in the CVS→CHI group. Neuroinflammation is being assessed in the CVS→CHI AC5216-treated mice. Taken together, these data suggest that increased neuroinflammation in CVS→CHI mice underlie their impaired cognition and increased anxiety.

Disclosures: K. St. Laurent-Ariot: None. **A. Fesharaki:** None. **J. Miyauchi:** None. **S.E. Tsirka:** None. **P.J. Bergold:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.04/DD4

Topic: C.09. Brain Injury and Trauma

Support: Otago Medical Research Foundation of New Zealand

Title: Post-treatment with melatonin prevents cerebral myelin and memory deficits in male rats exposed to repeated hypoxia during the equivalent of extreme prematurity

Authors: *D. E. OORSCHOT, S. NARAYANAN, L. GODDARD
Anat., Univ. Otago Sch. Biomed Sci., Dunedin, New Zealand

Abstract: The birth of an extremely premature baby (≤28 weeks of gestation) is costly to health systems. Children born extremely prematurely can experience repeated hypoxic injury to the brain and can develop memory and myelin deficits. In a new Sprague-Dawley rat model of repeated hypoxic brain injury during the equivalent of extreme prematurity, decreased O4-positive pre-oligodendroglia, decreased cerebral myelin and memory deficits were observed (Oorschot et al., 2013, *Journal of Neuroscience* 33:11863-11877). Yet, treatment with melatonin, which is safe in clinical neonatology and a known antagonist of the proposed biological mechanisms that generate a lower number of O4-positive pre-oligodendroglia, has not been investigated in terms of rescuing cerebral myelin or preventing memory deficits. Hence, the aim of this study was to investigate whether treatment with melatonin rescues memory and cerebral myelin. Postnatal day (PN) 1-3 Sprague-Dawley male rats were exposed to repeated hypoxia and divided into treatment groups of repeated hypoxia diluent-treated and repeated hypoxia melatonin-treated. The treatment commenced after the second repeated hypoxic exposure. A first cohort of rats (n = 4 pairs) were perfuse-fixed at PN14/15, and the left cerebral hemisphere was serially sectioned and then immunohistochemically stained with an antibody to myelin basic protein. The absolute surface area of myelin was then measured using stereological methods. After treatment with melatonin, there was a significant increase in the absolute surface area of myelin in the anterior commissure and the subcortical white matter (paired two-tailed Student's *t*-test, $p = 0.044$ and $p = 0.039$, respectively). There was also an increase in the absolute surface area of myelin in the corpus callosum after treatment with melatonin, but this increase was not statistically significant (paired two-tailed Student's *t*-test, $p = 0.124$). A second cohort of rats were tested for memory on the radial maze from PN35-45 (n = 10 melatonin-treated, n = 9 diluent-treated, n = 3 uninjured controls). The percentage of rats per day that achieved 100% success over 11 days on the radial arm maze was significantly different between the three groups (one-way ANOVA, $p = 0.006$). Post-hoc comparisons revealed a significant increase in the repeated hypoxia melatonin-treated rats and the control uninjured rats compared to the repeated

hypoxia diluent-treated rats ($p < 0.01$ and $p < 0.05$, respectively, Tukey's test, corrected for multiple comparisons). Thus, post-treatment with melatonin prevents cerebral myelin and memory deficits in male rats exposed to repeated hypoxia during the equivalent of extreme prematurity.

Disclosures: D.E. Oorschot: None. **S. Narayanan:** None. **L. Goddard:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.05/DD5

Topic: C.09. Brain Injury and Trauma

Title: Docosahexaenoic acid enhances the therapeutic potential of neural stem cell transplantation post traumatic brain injury

Authors: *F. H. KOBEISSY[1], N. RAMADAN[2], H. GHAZALE[2], H. DARWISH[3], W. ABOU KHEIR[4], J. SOUEID[2]
[1]Dept of Psychiatry, Univ. of Florida, Gainesville, FL; [2]Dept. of Biochem. and Mol. Genetics, Fac. of Medicine,, [3]Harriri school of Nursing, Fac. of Med., [4]Dept. of Anatomy, Cell Biol. and Physiological Sciences, Fac. of Medicine, American Univer, American Univ. of Beirut, Beirut,, Lebanon

Abstract: Traumatic Brain Injury (TBI) is a major cause of death and disability worldwide with 1.5 million people inflicted yearly. TBI is associated with severe post-TBI symptoms such as sensorymotor deficits. Several neurotherapeutic interventions have been proposed including drug administration as well as cellular therapy including neural stem cell (NSC)-based treatment. Among the proposed drugs utilized is docosahexaenoic acid (DHA), a poly-unsaturated fatty acid, exhibiting neuroprotective properties. In this study, we propose to combine an innovative intervention of neonatal NSCs transplantation in combination with DHA injections in order to ameliorate brain damage and promote functional recovery. Thus, NSCs derived from the subventricular zone of neonatal pups, cultured into neurospheres, were transplanted in an experimental controlled cortical impact mouse model of TBI. Effect of NSC transplantation was assessed alone and in combination with DHA administration. Motor deficits were evaluated using pole climbing and rotarod tests. Using immunohistochemistry, the effect of transplanted NSCs and DHA treatment was assessed by quantification of doublecortin (DCX), glial fibrillary acid protein (GFAP), ionized calcium binding adaptor molecule (IBA-1), and tyrosine hydroxylase (TH) markers. Combined NSC transplantation and DHA injections significantly attenuated TBI-induced motor function deficits (pole climbing tests), promoted neurogenesis coupled with an increase in glial reactivity at the cortical site of injury, and interestingly, with a marked increase in the number of dopaminergic neurons in the ventral tegmental area and the

substantia nigra. These data demonstrate that prior treatment with DHA may be a desirable strategy to improve therapeutic efficacy of NSC transplantation in TBI.

Disclosures: F.H. Kobeissy: None. **N. Ramadan:** None. **H. Ghazale:** None. **H. Darwish:** None. **W. abou Kheir:** None. **J. Soueid:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.06/DD6

Topic: C.09. Brain Injury and Trauma

Support: NIH Grant NS050465

Title: Functional brain network reveals enhanced rehabilitative potential by combining pharmacology and exercise after traumatic brain injury

Authors: *G. KRISHNA[1], Z. YING[1], A. PAYDAR[2], N. G. HARRIS[2], F. GOMEZ-PINILLA[1,2]
[1]Dept. of Integrative Biol. & Physiology, UCLA, Los Angeles, CA; [2]Dept. of Neurosurg., UCLA Brain Injury Res. Ctr., Los Angeles, CA

Abstract: Traumatic brain injury (TBI) often progresses to persistent disability accompanied with cognitive and psychiatric pathology. Although exercise has a rehabilitative value, its action on brain functional connectivity (fc) is poorly understood, which might limit efficacy. Brain fc is crucial for processing of information but it is disrupted by TBI therefore reducing outcome. Therefore, here we investigate how the concurrent application of exercise and pharmacology provided by 7,8-dihydroxyflavone (7,8-DHF, a neurotrophin receptor agonist) can stimulate rehabilitative training efficacy provided by exercise. Here we describe how 7,8-DHF and exercise influence resting-state functional MRI (rsfMRI) after TBI to characterize functional network properties. Adult male Sprague-Dawley rats (n= 5-6/group) were subjected to moderate fluid percussion injury (FPI) and received treatment with 7,8-DHF (5 mg/kg, i.p.), and half of them had access to voluntary wheel running for 2 consecutive weeks. rsfMRI was performed using a 7Tesla MRI scanner using a single-shot, echo-planar, gradient-echo sequence to measure BOLD contrast at 2h following the final treatment. We show that 7,8-DHF and exercise improved spatial memory responses, and activated TrkB signalling and cell metabolism post-injury. Initial assessment of global fc revealed that TBI decreased inter-hemispheric fc among hippocampus, sensory (S1), motor (M1), cingulate (Cg) regions of the cortex and caudate putamen (CP). The altered fc among these regions likely lead to inefficient spatial memory functioning among FPI animals. 7,8-DHF treatment and exercise increased fc ($P < 0.05$, uncorrected) in some of the same regions that were functionally disconnected after FPI. These preliminary findings highlight the efficacy of interventions to promote reorganization of

functional pathways, particularly, in networks located in regions implicated in cognitive and sensorimotor processing. Enhancement in fc may be an important biomarker after to predict success of the rehabilitative potential of exercise and 7,8-DHF interventions, and this would ultimately have important clinical implications.

Disclosures: G. Krishna: None. **Z. Ying:** None. **A. Paydar:** None. **N.G. Harris:** None. **F. Gomez-Pinilla:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.07/DD7

Topic: C.09. Brain Injury and Trauma

Support: ANR-Traumep-13-BSV4-0012-01

Fondation Gueules Cassées

Eranet Neuron III Acrobat

Title: Role of chloride homeostasis in post-traumatic depression

Authors: *E. GOUBERT[1,2], I. KHALILOV[1,2], M. ALTVATER[3], M. SCHAEFER[3], C. RIVERA[1,2,4], C. PELLEGRINO[1,2]
[1]INMED INSERM, Marseille, France; [2]Aix Marseille Univ., Marseille, France; [3]Dept. of Anesthesiol. and Res. Ctr. Translational Neurosciences, Mainz, Germany; [4]Helsinki University, Neurosci. Ctr., Helsinki, Finland

Abstract: Traumatic brain injury is one of leading cause of major depressive disorders, which represents a major public health concern. The identification of early neurological changes and whether they have later consequences on the depression onset represent critical issues. We have established and validate the controlled cortical impact rodent model of traumatic brain injury. Our current data suggest that there is early chloride homeostasis impairment leading to depressive-like behavior at later stages. Interestingly, an early treatment aiming at restoring chloride homeostasis during the first post-traumatic week significantly ameliorates the depression-like behavior at chronic stages. We will present result from experiments aiming to dissect out the mechanism of chloride homeostasis impairment and the settling up of depression. We highlight that post-traumatic dentate gyrus adult neurogenesis and GABAergic neurotransmission are key events in those processes. This will provide new insights on the early mechanisms underlying the onset of post-traumatic depressive disorders.

Disclosures: **E. Goubert:** None. **I. Khalilov:** None. **M. Altvater:** None. **M. Schaefer:** None. **C. Rivera:** None. **C. Pellegrino:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.08/DD8

Topic: C.09. Brain Injury and Trauma

Support: K12 HD27748-11

Title: IL-1RI blockade results in decreased cytokine expression and improved learning following TBI

Authors: ***B. TODD**, J. MAHONEY, P. FERGUSON, A. G. BASSUK, E. A. NEWELL
Univ. of Iowa, Iowa City, IA

Abstract: Neurologic injury from traumatic brain injury (TBI) is due to primary and secondary injury mechanisms. Neuroinflammation is one secondary injury mechanism felt to contribute to ongoing cell death following TBI. Interleukin-1 (IL-1) is a key regulator of neuroinflammation; IL-1α and IL-1β are the best-characterized cytokines of this family and signal through type I IL-1 receptor (IL-1RI). We aim to isolate the role of IL-1RI in TBI pathogenesis and evaluate the therapeutic potential of pharmacologic IL-1RI blockade using anakinra, recombinant IL-1RI antagonist. We use lateral fluid percussion injury model adapted to mice targeting moderate-severe TBI. C57BL/6J and global IL-1RI KO male mice (on C57BL/6J background) were used. In pharmacologic studies, systemic anakinra via IP injection was compared to saline control. Gene expression was evaluated by qPCR. Barnes maze testing was used to evaluate cognitive function 2 weeks post-FPI. Immunohistochemistry staining for IgG was used to evaluate blood brain barrier (BBB) breakdown. FPI resulted in a diffuse inflammatory cytokine response, with increased IL-1α, IL-1β, TNF and IL-6 expression in both focal (left parietal cortex, hippocampus) and remote regions (brainstem, cerebellum). IL-1RI ablation resulted in decreased IL-1β and IL-6 expression in remote regions (brainstem, cerebellum) 6 hours post injury, and decreased IL-1β and IL-6 expression in both focal (parietal cortex) and remote (brainstem) regions at 24 hours. Mice treated with anakinra, did not show altered inflammatory cytokine expression at 6 hours, but did show decreased IL-1β expression in the parietal cortex at 24 hours. Significant IgG immunoreactivity was present at 24hrs post-FPI and was maximal in peri-lesional cortex. Following FPI, anakinra treated mice showed improved learning in the Barnes maze compared to saline treated mice. Conclusions: FPI results in a diffuse inflammatory cytokine response, with increased expression of pro-inflammatory cytokines in focal and remote regions. Genetic IL-1RI ablation decreased the spread and hastened resolution of CNS inflammatory cytokine expression following FPI. Pharmacologic IL-1RI blockade did not

prevent the spread of inflammatory cytokine expression, but did hasten the resolution of IL-1β in the parietal cortex. This focal pharmacologic effect may be due to regional differences in CNS drug penetration as cytokine modulation mirrored area of maximal BBB breakdown. Pharmacologic IL-1RI blockade following TBI is a potential therapeutic strategy but dosing and delivery methods need to be optimized for maximal beneficial effect. Support: K12 HD27748-11

Disclosures: B. Todd: None. **J. Mahoney:** None. **P. Ferguson:** None. **A.G. Bassuk:** None. **E.A. Newell:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.09/DD9

Topic: C.09. Brain Injury and Trauma

Support: NIH Grant NINDS 1DP2HD084067

NIH Grant DP2HD084067-01S1

Title: Antibody fragment phage display as a biomarker discovery tool for traumatic brain injury

Authors: *B. I. MARTINEZ, G. MOUSA, S. STABENFELDT
Arizona State Univ., Tempe, AZ

Abstract: Introduction: Traumatic brain injury (TBI) affects 1.7 million people in the United States each year, costing billions of dollars in medical care expenses. The temporal nature of the secondary injury that progresses after the initial insult contributes to a complex pathology that confounds efforts to develop TBI diagnostic and therapeutic modalities. Therefore, it is critical to identify unique neural injury biomarkers at distinct injury phases to improve prognosis and intervention efforts. Here, we combined antibody fragment phage display with the potency of next generation sequencing (NGS) analysis as a biomarker discovery tool for TBI at the acute and subacute post-injury phases. We postulated that unique sets of antibody fragments that recognize neuropathology at each phase following injury would be uncovered. **Methods:** Animal studies were conducted in accordance to protocols approved by ASU IACUC. Adult C57Bl/6 mice were anesthetized and subjected to either unilateral controlled cortical impact (CCI) injury or a sham surgery (n=3 per group/timepoint). Animals were randomly assigned to either the acute (1 day) or subacute (7 days) post-injury/sham group and intravenously injected with 100 µL of human domain antibody (dAb) phage library at a concentration of 10^9-10^{12} phage. Bound phage were eluted from the brain and control tissue pools and quantified with titers. Phage eluted from injured ipsilateral brain tissue were amplified for the second biopanning round. Phage amplified from recovered tissues and control libraries were sequenced using Illumina's MiSeq

2x300 module. Analysis was completed using the FASTAptamer Toolkit package and Bioconductor for R. **Results:** NGS analysis revealed that each tissue and control library sample yielded 7000-12,000 antibody fragment sequences. Looking specifically at sequences from the injured ipsilateral brain tissue, there were 109 identical sequences in both the acute and subacute injured brain tissue. 64 of 109 sequences appeared more frequently in the subacute group than the acute group. A similar comparison between sham and injury groups of ipsilateral tissue yielded less than 90 shared sequences, the majority of which occurred with markedly decreased frequency within injury groups. **Discussion:** Our *in vivo* phage display biopanning process is sensitive to the robust temporal effects of neural injury, thus demonstrating its efficacy as a biomarker discovery tool for TBI. Further studies will isolate unique candidate biomarkers for each timepoint and further characterize their specificity to distinct phases of neural injury.

Disclosures: B.I. Martinez: None. **G. Mousa:** None. **S. Stabenfeldt:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.10/DD10

Topic: C.09. Brain Injury and Trauma

Title: Improving outcomes: An emerging role for zinc chelation and pH in traumatic brain injury

Authors: *Z. WANG[1], R. TIAN[2]
[1]Biomed. Sci., Ohio Univ. Dept. of Biomed. Sci., Athens, OH; [2]Neurosurg., Beijing Tiantan Hospital, Capital Med. University, Beijing Neurosurgical Inst., beijing, China

Abstract: Traumatic brain injury (TBI) occurs when an external mechanical force blows the brain, resulting in brain dysfunction. Mild traumatic brain injury may cause temporary dysfunction of brain cells. More serious traumatic brain injury can result in bruising, torn tissues, bleeding and other physical damage to the brain that can result in long-term complications or death. According to the data from CDC, the number of reported TBI cases is over 2 million in 2010 in the United States, which becomes a major cause of death and disability, especially in children and young adults. The major causes of TBI are falls, motor vehicle traffic injuries, and strikes. Numerous of previous works demonstrated that TBI-induced excessive zinc release from neurons, resulting in neuronal death and damage. This damage might be caused by the formation of zinc accumulation. Furthermore, some researchers indicate that following an acute brain injury, cerebral blood flow and cerebral metabolism of patients will be disturbed, resulting in brain pH decrease in the beginning, followed by latterly pH increasing. Decreased pH in the brain will lead to zinc release from loosely bound proteins, and later pH increase will promote the production of zinc precipitate. In the present study, we investigated the effect of zinc chelation on TBI-induced brain infraction and edema in a mice model. We proposed a strategy to

improve TBI outcomes through reducing the formation of zinc precipitate. Both acidic solution (pH~4) and zinc chelator, Ca-EDTA, was directly applied to the brain damage region. TTC staining was used to measure brain infarction and edema levels. Results showed that both acidic solution and zinc chelator showed a benefit effect on TBI outcomes. Both acidic condition and zinc chelation exhibited a neuroprotective effect on TBI-induced brain damage through reducing brain infarct volumes and edema areas. Zinc chelation and acidic solution showed a more neuroprotective effect on decreasing infarct volume than that of edema levels. Furthermore, the treatment with zinc chelator presented more powerful effect than that of the acidic solution on anti-infarct volume but anti-edema formation after TBI.

Disclosures: Z. Wang: None. **R. Tian:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.11/DD11

Topic: C.09. Brain Injury and Trauma

Title: NYX-2925 regulates tau dynamics in rat cortical tissues

Authors: *L. P. CACHEAUX[1], K. LEADERBRAND[1], M. SCHMIDT[1], E. COLECHIO[1], J. S. BURGDORF[1,2], R. A. KROES[1,2], J. R. MOSKAL[1,2]
[1]Aptinyx Inc., Evanston, IL; [2]Falk Ctr. for Mol. Therapeutics, McCormick Sch. of Engin., Northwestern Univ., Evanston, IL

Abstract: Aptinyx has developed a novel class of small molecule N-Methyl-D-Aspartate (NMDA) receptor modulators with broad applicability across neurologic and psychiatric disorders. It is a robust cognitive enhancer that binds with high affinity to all four NR2 subtypes and rapidly enhances synaptic plasticity processes associated with learning and memory. Oral administration of NYX-2925 (1 mg/kg, 1-24 hr post-injury) ameliorates learning deficits in a rat model of traumatic brain injury (TBI). In both humans and rodent models, TBI has been shown to result in both cognitive deficits and altered tau phosphorylation. Because NMDAR activation regulates phosphorylation of endogenous tau, the effects of NYX-2925 on tau expression and phosphorylation were investigated as a putative mechanism underlying its therapeutic efficacy in TBI. Naïve rats were treated with either vehicle (0.5% CMC in saline; 1 ml/kg, PO) or NYX-2925 (1 mg/kg, PO), and medial prefrontal cortex (mPFC) proteins were isolated at 15, 30, 60 min, and 24 hr post-dosing. Both total and phosphopeptides enriched by HILIC fractionation were analyzed using nano-LC MS/MS analysis. This unbiased proteomic approach identified significant time-dependent changes in total tau and tau phosphorylation at 16 sites, primarily serine and threonine residues. These changes were confirmed via Western blot analysis in rat primary cortical cultures treated with NYX-2925. This work provides evidence that targeting

NMDAR-associated modulation of tau phospho-dynamics is a promising therapeutic strategy for TBI.

Disclosures: L.P. Cacheaux: A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **K. Leaderbrand:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **M. Schmidt:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **E. Colechio:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **J.S. Burgdorf:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **R.A. Kroes:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **J.R. Moskal:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc..

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.12/DD12

Topic: C.09. Brain Injury and Trauma

Support: NIH Grant RO1 NS 12542

NSF Grant EEC-1028725

NIH Grant 1F30NS100253-01

Title: Modulation of single-neuron and network activity in motor cortex by clinically realistic transcranial direct current stimulation in non-human primates

Authors: *A. R. BOGAARD[1,2], H. M. BOYD[2], A. MORSE[2], S. ZANOS[3], E. E. FETZ[2]
[1]Med. Scientist Training Program, [2]Physiol. and Biophysics, Univ. of Washington, Seattle, WA; [3]Feinstein Inst. for Med. Res., Manhasset, NY

Abstract: Transcranial direct current stimulation (tDCS) is a popular brain stimulation technique used in human trials because it is noninvasive and simple to implement. Despite its popularity, tDCS is controversial because results are variable and the mechanism of action is not well understood. The reason for this is twofold: animal models of tDCS do not resemble that used with patients, and studies with humans generally use secondary outcomes to infer physiological effects. In the present study, we set out to test the prevailing theories underlying tDCS by directly measuring effects on brain activity in a non-human primate model of tDCS which closely matches that used in humans. Recordings of single unit activity (SUA) and local field potentials (LFP) were obtained from two macaca nemestrina monkeys by 96 channel microelectrode arrays implanted over the forelimb area of left motor cortex (MC). tDCS was delivered to the scalp by saline soaked sponge electrodes in a unilateral montage to target anodal or cathodal polarization to the area of MC being recorded, and intensity ranged from low in human studies (0.027 mA/cm^2), to higher than that used in humans (0.5 mA/cm^2). Each monkey was trained in a visuomotor target tracking task in which the position of a cursor was controlled by a 2-axis manipulandum that registered isometric wrist torques. During each trial, the monkey was cued to move the cursor to one of eight targets by contracting his forearm muscles. Each trial was followed by a period of quiet sitting (0.5-2 sec) to compare effects of tDCS on active and resting cortex. First, we looked for evidence that anodal and cathodal tDCS have opposite modulatory effects on SUA. We recorded over 3000 neurons from 90 experiments (29 anodal, 28 cathodal and 33 control), and found a modest but significant increase in median firing rate during anodal tDCS. This increase in firing was positively correlated with current intensity and was significant only for currents over 0.13 mA/cm^2, and persisted after tDCS was turned off. Interestingly, cathodal tDCS produced no significant change in firing at any current intensity. Second, we looked for changes in the baseline LFP beta power during quiet sitting. We observed a significant increase in beta power during cathodal tDCS, and a smaller, but significant, decrease in beta power during anodal tDCS. Lastly, we found no evidence that tDCS affected the shape of motor directional tuning or spike-field coherence. As far as we know, this is the first direct demonstration of how clinical-type tDCS modulates neuron firing and network activity.

Disclosures: A.R. Bogaard: None. **H.M. Boyd:** None. **A. Morse:** None. **S. Zanos:** None. **E.E. Fetz:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.13/DD13

Topic: C.09. Brain Injury and Trauma

Support: contract 13-8A from Kentucky Spinal Cord and Head Injury Research trust

Title: Pre-treatment with the macrolide antibiotic azithromycin increases tissue sparing after cortical contusion brain injury in rats

Authors: *K. B. HUBER, D. HOPKINS, J. R. PAULY
Pharmaceut. Sci. PhD program, Univ. of Kentucky, Lexington, KY

Abstract: Traumatic brain injury involves a mechanical injury followed by secondary biochemical cascades that propagate further damage including production of reactive oxygen species, and release of pro-inflammatory factors from resident innate immune cells. No pharmacological therapeutics have translated from preclinical models to clinical trials, highlighting the need for further research. Neuroinflammation has gained prominence as an important mediator of secondary damage after injury. Azithromycin (AZM) treatment has been shown to increase M2 polarization and decrease lesion volumes/motor deficits in models of spinal cord injury and stroke (Zhang et al., 2015; Amantea et al., 2016). The purpose of this study was to determine if AZM pretreatment would promote neuroprotection and enhance behavioral recovery following experimental cortical contusion brain injury. Adult male Sprague-Dawley rats were administered 160mg/kg of AZM daily, via oral gavage, for once daily three days prior to cortical contusion injury (2.0mm depth, 3.5m/s, 2.5mm tip diameter, 500ms dwell time) and once 15 minutes post-injury. Morris Water Maze (MWM) testing was conducted for 4 days (4 trials per day) starting on day 4 post-injury to assess spatial memory. There was no significant difference in performance between AZM and vehicle treated animals in the MWM. Following behavioral testing, animals were euthanized and the brains were prepared for a cortical tissue sparing analysis, and assessment of neuroinflammation using [^{3}H]-PK-11195, which binds to the translocator protein 18kDa (TSPO). In spite of the lack of effects on cognitive behavior, AZM treatment was associated with significant cortical tissue sparing (89.0 ± 4.8 percent spared) compared to vehicle treated animals (78.4 ± 4.8 percent spared; $p<0.0001$). Neuroinflammation, as assessed by PK-11195 binding, was significantly increased in all animals with head injuries, but there were no robust differences between animals treated with vehicle and AZM. We conclude that AZM exhibited significant neuroprotection against overt tissue loss, but more studies will need to be conducted to elucidate the nature of its effect on cognitive deficits and inflammatory signaling post-injury.
Supported by Contract 13-8A from the Kentucky Spinal Cord and Head Injury Research Trust.

Disclosures: K.B. Huber: None. **D. Hopkins:** None. **J.R. Pauly:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.14/DD14

Topic: C.09. Brain Injury and Trauma

Support: NINDS-NIH R21 NS093268

NIH UL1TR000457

Title: L-DOPA induces reversibility of dopamine presynaptic deficit in minimally conscious state patients following traumatic brain injury

Authors: *E. A. FRIDMAN[1], J. R. OSBORNE[2], N. D. SCHIFF[1]
[1]Weill Cornell Med., New York, NY; [2]Mem. Sloan Kettering Cancer Ctr., New York, NY

Abstract: Minimally conscious states (MCS) following severe TBI express evidence of conscious awareness or goal-directed behaviors. Pharmacological stimulation can markedly improve some MCS patients. We previously demonstrated the use of [^{11}C]raclopride-PET a novel biomarker to identify a presynaptic dopamine deficit affecting the mesolimbic and mesothalamic pathways in MCS (Fridman et al. SfN 2015). However, no studies have provided direct measurements to test the reversibility of a presynaptic dopaminergic deficit in MCS. Here, we compared the availability of dopamine D2-like receptors in five MCS patients at a baseline, induced after prior dopamine reuptake blockade with dextroamphetamine (AMPH), and after a single dose of 375mg of $_{L}$-DOPA. We manually defined in high-resolution MRI the following structures: the ventral striatum (VST), associative striatum (AST) and sensorimotor striatum (SMST); and, the central thalamus (c-TH). We analyzed the signal using the kinetic SRTM2 with cerebellum as reference tissue to extract binding potential nondisplaceable (BP_{nd}) from ROIs. The mean delta % change (Δ BP_{nd}) between pre- and post-AMPH and between pre- and post-$_{L}$-DOPA were calculated and analyzed using ANOVA (Δ BP_{nd} as dependent variables and condition as independent variable in each ROI). Results showed that despite persistent deficit in the presynaptic dopaminergic neurons after administration of AMPH, 4 out of 5 MCS patients could increase dopamine release and reverse the deficit after a single dose of $_{L}$-DOPA. Specifically, when comparing the mean Δ BP_{nd} in MCS post-AMPH to those obtained post-$_{L}$-DOPA we identified a significant reduction in AST, SMST and c-TH [AST: AMPH =-9.9 ± 2.9, $_{L}$-DOPA =-23.0 ± 3.7; F= 7.9, p =0.01; SMST: AMPH =-7.0 ± 3.5, $_{L}$-DOPA =-20.9 ± 3.9; F= 6.4, p= 0.02; c-TH: AMPH =-0.1 ± 6.1, $_{L}$-DOPA =-23.7 ± 7.6; F= 6.1, p= 0.03]. We interpret findings as demonstrating that administration of exogenous $_{L}$-DOPA restores dopamine biosynthesis via bypassing the first step of the process; thus, our observations suggest that a deficiency of the enzyme tyrosine hydroxylase is present in post-TBI MCS, accounting for the inability of blocking the dopamine transporter reuptake alone to increase dopamine availability.

Disclosures: E.A. Fridman: None. **J.R. Osborne:** None. **N.D. Schiff:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.15/DD15

Topic: C.09. Brain Injury and Trauma

Support: George Mason University OSCAR Program

Title: The enhancement of neuroscience educational tools for subjects with traumatic brain injury

Authors: J. ORTIZ[1], F. BERLIN[1], *G. LEWIS[2]
[2]Col. of Sci., [1]George Mason Univ., Fairfax, VA

Abstract: The knowledge divide that separates non-scientists, health-care professionals, and research investigators has led to inadequate patient education within the medical field. Often, the only materials patients receive are relatively complex medical packets that are difficult for non-medical professionals to understand. This problem is made worse for patients who suffer Traumatic Brain Injuries (TBIs). These patients often suffer cognitive deficits and have difficulties with reading comprehension. Here we investigate the efficacy of a novel program for patient education in TBI subjects. In this study, TBI patients participated in a series of 3 engaging lesson plans that were delivered in a one-on-one format. Lesson plans topics focused on 1) the central and peripheral nervous system, 2) the regenerative properties of the nervous system, and 3) the biological properties of Traumatic Brain Injury. Lesson plans were delivered by a personal tutor in a one-on-one setting, where patients had the opportunity to ask questions and were free to interact with the tutor. Visual aids were provided in the form of white-board drawings and a 1-page learning map, which clearly summarized the important concepts of each lesson on a single page. Pre and post lesson quizzes and surveys were conducted to assess learning and confidence. Pre and post study interviews were conducted to determine the perceived value of the lessons to participants and to collect qualitative data about participant's injuries and experiences. Our data suggests this educational program improved patient's knowledge and confidence in understanding basic neuroscience and TBI concepts. Improvements in patient knowledge and confidence could increase the quality of communication between patients, doctors and caregivers. This may ultimately aid in decisions regarding the care, treatment, and life-management of individuals who have suffered TBI.

Disclosures: **J. Ortiz:** None. **F. Berlin:** None. **G. Lewis:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.16/DD16

Topic: C.09. Brain Injury and Trauma

Support: US Army Combat Casualty Care Research Program H_026_2014_WRAIR

Title: Evidence of mitochondrial energy dysfunction following penetrating ballistic-like brain injury

Authors: *J. D. PANDYA, Y. DENG-BRYANT, X. YANG, L. Y. LEUNG, D. A. SHEAR
Brain Trauma Neuroprotection and Neurorestoration (BTNN), Walter Reed Army Inst. of Res. (WRAIR), Silver Spring, MD

Abstract: Mitochondria play a pivotal role in the development of secondary pathophysiology and subsequent neuronal cell death following traumatic brain injury (TBI). Under normal circumstances, brain consumes glucose as the preferred energy source for adenosine triphosphate (ATP) production over ketones. However, the bioenergetics profile of glucose intermediate metabolites and ketones have not been individually compared following TBI. We evaluated the injury-induced alterations in brain mitochondrial bioenergetics, enzyme activities and coenzyme contents in the penetrating ballistic-like brain injury (PBBI) model of TBI. Adult male rats were subjected to either 10% unilateral PBBI or Sham craniotomy (n=5 per group). At 24 hours post-injury, mitochondria were isolated from pooled brain regions (i.e. frontal cortex and striatum) of the ipsilateral hemisphere. Mitochondrial bioenergetics parameters were measured in the presence of metabolic substrates pyruvate+malate (PM), glutamate+malate (GM), succinate (Succ) and β-hydroxybutyrate+malate (BHBM). Mitochondrial electron transport chain (ETC) complex enzyme activity i.e. NADH dehydrogenase (Complex I), succinate dehydrogenase (Complex II) and cytochrome c oxidase (Complex IV) were evaluated in isolated mitochondria of the Sham and PBBI groups. Additionally, the tricarboxylic acid (TCA) cycle dehydrogenases enzyme activity i.e. pyruvate dehydrogenase complex (PDHC), alpha-ketoglutarate dehydrogenase complex (α-KGDHC), glutamate dehydrogenase (GDH); and mitochondrial coenzyme contents were quantitated. Significant reductions in ATP synthesis were detected in PBBI group with PM (-43%), Succ (-50%) and BHBM (-44%) as respiratory substrates Vs Sham. When compared ATP synthesis rates of four substrates within Sham or PBBI group; the PBBI group had a distinct substrate utilization pattern for ATP synthesis. In the PBBI group, mitochondrial Complex I (-50%) and Complex IV (-80%) enzyme activity were significantly reduced; whereas Complex II activity was comparable to Sham. The PDHC (-45%) and GDH (-50%) activity were significantly reduced in the PBBI group as compared to Sham; whereas no significant differences were noted in α-KGDHC activity between two groups. Additionally, mitochondrial coenzyme contents (i.e. FAD and NAD) were significantly decreased (-25% to -35%) in PBBI Vs Sham group. Collectively, PBBI leads to an overall reduction and shift in the metabolic activity that may be caused by significant reductions in brain mitochondrial ETC and TCA cycle enzyme activities. These results provide a strong basis for the use of “alternative biofuels” following brain trauma.

Disclosures: J.D. Pandya: None. **Y. Deng-Bryant:** None. **X. Yang:** None. **L.Y. Leung:** None. **D.A. Shear:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.17/DD17

Topic: C.09. Brain Injury and Trauma

Support: Boston Children's Hospital Translational Research Program

Title: Magnetic resonance spectroscopy detects early, regional glutathione decrease and restoration by N-acetylcysteine following traumatic brain injury in rats

Authors: *R. M. GUERRIERO[1,2], M. HAMEED[2], N. W. HODGSON[2], B. ROWLAND[3], P. L. PEARL[2], B. E. KOSOFSKY[4], T. K. HENSCH[5], A. LIN[3], A. ROTENBERG[2]
[1]Neurol., Washington Univ. Sch. of Med., Saint Louis, MO; [2]Boston Children's Hosp., Boston, MA; [3]Brigham And Women's Hosp., Boston, MA; [4]Ped Neurol. Div., Weill Cornell Med. Col., New York, NY; [5]Boston Children's Hosp. & Harvard Univ., Boston, MA

Abstract: Objective: To evaluate magnetic resonance spectroscopy (MRS) as an *in vivo* tool to detect early biomarkers of injury in a rat traumatic brain injury (TBI) model. **Background**: TBI leads to glutamate excitotoxicity and oxidative stress, which lead to neuronal injury and death. Rising extracellular glutamate levels inhibit the transport of cystine into astrocytes via the glutamate/cystine antitransporter. This inhibition decreases the amount of cystine available for conversion to cysteine, the rate-limiting precursor for glutathione (GSH) synthesis. A high reduced:oxidized GSH ratio is essential for maintaining the neuronal redox state and mitigating oxidative stress after TBI. In the days after injury, parvalbumin-positive inhibitory interneurons are injured preferentially in the post-traumatic cortex, as they are highly metabolically active and particularly sensitive to oxidative stress. An *in vivo* tool to serially measure regional GSH remains an unmet need. **Methods:** Adult male rats underwent a rapid fluid percussion injury. One group of rats was treated with N-acetylcystine (NAC) for the first week post-injury. The three groups were: sham, TBI/NAC (T/N), and TBI/vehicle (T/V). Animals were studied by MRS on a 7T Bruker rodent MRI 7 days after injury with 2 x 2 x 2 mm voxels using PointRESolved Spectroscopy (PRESS) localization, water suppression, Fast Automatic Shimming Technique by Mapping Along Projections (FASTMAP) with 256 repetitions and regions of interest over lesional and contralesional cortex. MRS data was analyzed using linear combinations model (LCmodel, Provencher). Metabolite measures with Cramer-Rao lower bound of less than 20% standard deviation were analyzed. Rats were sacrificed 7 days after TBI for tissue processing for high performance liquid chromatography (HPLC) to measure GSH and its metabolites. **Results**: MRS on day 7 following injury showed that both N-Acetylaspartic acid (NAA), a marker of neuronal health and GSH levels were significantly ($p < 0.05$) decreased in T/V animals (N=8) compared to sham (N=4). Both NAA and GSH levels were restored to

normal levels following NAC treatment (N = 13) compared to the T/V group ($p< 0.01$) and were not different from the sham group. These data corresponded to a decrease in the ratio of reduced GSH to its oxidized form in lesional cortex at Day 7 ($p < 0.01$) and restored GSH levels in T/N treated animals ($p < 0.05$) as measured by HPLC. **Conclusion**: Serial MRS measures detect pathologic alterations in GSH homeostasis from time of TBI through the first week following injury, which corresponds to neuronal loss. NAC treatment restores GSH levels as measured by MRS and HPLC and appears to be neuroprotective.

Disclosures: R.M. Guerriero: None. **M. Hameed:** None. **N.W. Hodgson:** None. **B. Rowland:** None. **P.L. Pearl:** None. **B.E. Kosofsky:** None. **T.K. Hensch:** None. **A. Lin:** None. **A. Rotenberg:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.18/DD18

Topic: C.09. Brain Injury and Trauma

Support: NS093920

Title: Small molecule inhibition of p38α-mediated neuroinflammatory response to traumatic brain injury

Authors: *J. M. MORGANTI[1], D. M. WATTERSON[2], L. J. VAN ELDIK[1]
[1]Univ. of Kentucky, Lexington, KY; [2]Northwestern Univ., Chicago, IL

Abstract: Traumatic brain injury (TBI) initiates a multitude of cellular responses in the brain following the primary injury. Aberrant neuroinflammatory signaling cascades are one of these hallmark responses. Consistently, dysregulation of neuroinflammation has been linked with propagating neurodegenerative sequelae following TBI. Of principal interest is the role of p38α mitogen activated protein kinase (MAPK) in regulating inflammatory-linked responses to TBI. In the current study, we examined the brain's acute and subacute inflammatory response to TBI after treatment of mice with a novel, brain-penetrant, small molecule p38α inhibitor, MW150. Focal TBI was reproduced using the controlled cortical impact method. Three post-injury time points were examined; 1 day, 3 day, and 7 day. For each time point, three groups were examined; sham-vehicle (saline), TBI-vehicle, and TBI-MW150 (5mg/kg). Saline or MW150 was administered at 3 and 6 hours after injury and daily (3 and 7 day groups only) thereafter. At the prescribed interval, animals were euthanized for tissue collection to examine gene expression via NanoString. Current results suggest that the post-injury timing of p38α inhibition is critical for remediating injury-induced inflammatory sequelae.

Disclosures: **J.M. Morganti:** None. **D.M. Watterson:** None. **L.J. Van Eldik:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.19/DD19

Topic: C.09. Brain Injury and Trauma

Support: NIH-NINDS 5R01 NS083405

NIH-NINDS 5R01 NS084857

NIH-NINDS F30 NS096876

Title: Neuroprotective strategies following severe controlled cortical impact traumatic brain injury: Lipid peroxidation-derived neurotoxic aldehyde scavenging and inhibition of mitochondrial permeability transition

Authors: *J. R. KULBE, ***J. R. KULBE**, I. N. SINGH, J. A. WANG, E. D. HALL
Spinal Cord and Brain Injury Res. Ctr., Univ. of Kentucky, Lexington, KY

Abstract: There are currently no FDA-approved pharmacotherapies able to prevent the devastating neurological deficits caused by traumatic brain injury (TBI). Following injury mitochondria buffer increases in intracellular Ca^{++}. However, an increase in intra-mitochondrial Ca^{++} leads to mitochondrial generation of reactive oxygen and nitrogen species (ROS/RNS), which initiate lipid peroxidation (LP) of cellular and organelle membranes, particularly mitochondrial membranes. LP is terminated upon formation of LP-derived neurotoxic aldehydes, which covalently bind cellular and mitochondrial proteins, exacerbating mitochondrial dysfunction and enhancing Ca^{++} induced opening of the mitochondrial permeability pore (mPTP), resulting in loss of ATP production and extrusion of Ca^{++} back into the cytosol where it can initiate a downstream cascade of calpain activation, cytoskeletal spectrin degradation, neurodegeneration and neurologic impairment. Therefore, mitochondria are promising therapeutic targets following TBI. Promising strategies of mitochondrial protection include scavenging of LP-derived neurotoxic aldehydes and inhibition of mPTP. Therefore, combining aldehyde scavenging with mPTP inhibition may enhance neuroprotection. Phenelzine (PZ), an FDA-approved monoamine oxidase (MAO) inhibitor used clinically as an anti-depressant, is capable of scavenging neurotoxic aldehydes. Cyclosporine A (CsA), an FDA-approved immunosuppressant, is capable of inhibiting mPTP. This is the first study to evaluate the neuroprotective effects of a continuous 72h subcutaneous infusion, representative of peak LP and mitochondrial dysfunction, of PZ, CsA, or PZ+CSA following a severe controlled cortical impact injury in rats. Results for the first dosing paradigm evaluated (PZ: 10mg/kg s.c. 15min

post-injury + 10mg/kg/day/72h; CsA: 20mg/kg i.p. 15min post-injury + 10mg/kg/day/72h) indicate that while PZ is able to maintain respiratory control ratio, a general measure of mitochondrial health, compared to sham, neither CsA nor PZ+CsA is able to improve mitochondrial respiratory function. However, PZ and CsA, but not the combination PZ+CSA, were able to attenuate modification of mitochondrial proteins by LP-derived neurotoxic aldehydes. However, it is premature to abandon the combination of aldehyde scavenging and inhibition of mPTP for neuroprotection following TBI. Therefore, additional outcome measures, including spectrin degradation and attenuation of cellular oxidative damage will be evaluated. Furthermore, the effects of MAO inhibition on mitochondrial function following TBI will need to be assessed.

Disclosures: **J.R. Kulbe:** None. **I.N. Singh:** None. **J.A. Wang:** None. **E.D. Hall:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.20/DD20

Topic: C.09. Brain Injury and Trauma

Support: NIH R01 HL071568

Title: Therapeutic hypothermia promotes cerebral blood flow recovery and brain homeostasis after resuscitation from cardiac arrest in a rat model

Authors: *Q. WANG, P. MIAO, H. MODI, S. GARIKAPATI, R. KOEHLER, N. THAKOR
Johns Hopkins Univ., Baltimore, MD

Abstract: ***Aim***: Laboratory and clinical studies has demonstrated that therapeutic hypothermia (TH), when applied as soon as possible after resuscitation from cardiac arrest (CA), results in better neurological outcome. This study aimed to test hypothesis that TH would promote cortical cerebral blood flow (CBF) restoration and maintenance after return of spontaneous circulation (ROSC) from CA.
Methods: Twelve Wistar rats resuscitated from 7 minutes of asphyxial CA were randomized into two groups: hypothermia group (7H, n=6), treated with mild TH (33~34°C) immediately after ROSC; and normothermia group (7N, n=6, 37.0±0.5°C). We recorded multiple parameters including mean arterial pressure, CBF via laser speckle contrast imager and neural activities via electroencephalogram (EEG). The neurological outcomes were evaluated using Information quantity (IQ) entropy of EEG and neurological deficit scale (NDS).
Results: TH promoted consistently stable CBF near baseline in 7H group compared to that of the 7N group (the average CBF during first 30-minute post ROSC of two groups was 7H vs 7N as 91.4% ± 0.45 % vs 76.7% ± 0.6 %, $p<0.01$). Rats in the 7H group showed significantly better IQ

scores by 10 minutes after ROSC and better NDS scores at 4 and 24 hours (55.0 ± 1.83 in 7H vs 49.3 ± 1.9 in 7N at 4h, $p<0.05$; 70.0 ± 2.62 in 7H vs 60.0 ± 2.65 in 7N at 24h, $p<0.05$; best score=80).
Conclusion: TH facilitates restoration of CBF back to baseline levels after asphyxial CA and is associated with the restoration of brain electrical activity and improved outcome.

Disclosures: Q. Wang: None. **P. Miao:** None. **H. Modi:** None. **S. Garikapati:** None. **R. Koehler:** None. **N. Thakor:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.21/DD21

Topic: C.09. Brain Injury and Trauma

Support: Dept of Veterans Affairs

Title: Peripheral neurotrophic factor signaling with chronic mild traumatic brain injury

Authors: *M. W. MCNERNEY, V. DARCY, L.-L. WU, D. WALTZMAN, G. SWAMINATH, M. YUTSIS, O. HARRIS, J. ASHFORD, J. YESAVAGE, A. SALEHI, M. ADAMSON
VA, Palo Alto, CA

Abstract: An estimated 15.2% to 22.8% of returning service members suffer from mild traumatic brain injury (mTBI), with many experiencing chronic post-concussive symptoms. Common symptoms include memory problems or cognitive impairments, fatigue, headache, and loss of emotional regulation. Immediately following injury, there is a release of numerous neurotransmitters and signaling factors that may also play an important role in chronic mTBI etiology. Brain-derived neurotrophic factor (BDNF) is one of these molecules, and is thought to be involved in neuronal recovery and regrowth following injury. However, research on its role in chronic mTBI is limited. Here, we measured plasma BDNF levels in 47 Veterans (mean age 44.5 years, SD = 13.03) with and without a history of mTBI. We found that even years following injury, there is a significant positive correlation between injury severity and circulating BDNF levels. Neither BDNF levels nor injury severity were related to overall hippocampal volume, but there was a relationship with the BDNF precursor, proBDNF, and the val66met single nucleotide polymorphism. Mechanisms for this increase in BDNF in terms of injury severity and recovery and genotype will be discussed.

Disclosures: M.W. McNerney: None. **V. Darcy:** None. **L. Wu:** None. **D. Waltzman:** None. **G. Swaminath:** None. **M. Yutsis:** None. **O. Harris:** None. **J. Ashford:** None. **J. Yesavage:** None. **A. Salehi:** None. **M. Adamson:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.22/DD22

Topic: C.09. Brain Injury and Trauma

Support: Brain Tumour Foundation of Canada

Schulich

Cancer Research Society

Lawson Health Research Insitute

Mitacs

Canadian Institute for Health Research

Ontario Graduate Scholarships

Title: Patient-derived glioblastoma cells are susceptible to intratumoral modulation therapy both in 2D and neurosphere culture

Authors: *A. DEWEYERT[1], A. DI SEBASTIANO[1], H. XU[2], C. DE OLIVEIRA[1], E. WONG[3], S. SCHMID[1], M. HEBB[2,1]
[1]Anat. and Cell Biol., [2]Clin. Neurosci., [3]Med. Biophysics, Univ. of Western Ontario, London, ON, Canada

Abstract: Introduction: Glioblastoma (GBM) is the most common and aggressive primary brain tumor in adults. Advances in electrotherapy offer new promise for GBM but the potential of this putative treatment modality has not yet been realized. Our group has been pioneering an innovative strategy called Intratumoral Modulation Therapy (IMT) which delivers pulsed electric current or fields directly into GBM tissues. IMT produces caspase-dependent apoptosis of GBM cells without significant impact on post-mitotic neurons. The goal of this work was to characterize the anti-tumor impact of a spectrum of IMT parameters and to define the resultant IMT treatment fields using in vitro and in vivo GBM models. **Materials and Methods:** Patient-derived and immortalized GBM cells were treated with IMT using an established in vitro model (Xu et al 2016). We have also established a syngeneic rodent GBM model for in vivo evaluation of IMT. Waveform generators were used to deliver sinusoidal or square waves pulses with varying frequency (100 Hz- 200 kHz) and amplitudes (4-10 V) directly to GBM cells or tumors. Pilot studies with various cancer cell lines known to cause brain metastases were also screened for susceptibility to IMT. Cell viability was assessed with activated caspase-3 labeling, 3-(4,5-

dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) viability assay and flow cytometry with Annexin V and propidium iodide detection. Image-based computer modeling was performed using COMSOL software to predict field strength and dimensions, and to reconstruct IMT treatment fields using various electrode configurations and stimulation parameters. **Results:** The MTT assay revealed a significant loss of metabolic viability in patient derived and rat GBM cells, as well as other cancer cell lines treated with various electrical permutations of IMT compared to the respective sham conditions *in vitro*. Activated caspase-3 was elevated and consistent with flow cytometry that demonstrated increased expression of apoptotic markers in treated GBM cells. To date *in vitro* measurements within our IMT model have corroborated early mathematical predictions of field intensity and dimensions generated by COMSOL. These in vitro methods have been translated successfully to the GBM animal model. **Discussion and Conclusions:** This data supports further studies to reveal the potential of IMT as a putative new treatment strategy for GBM and possibly other forms of brain tumors. Advances in field prediction mapping and optimization of IMT hardware and stimulation parameters are needed.

Disclosures: **A. Deweyert:** None. **A. Di Sebastiano:** None. **H. Xu:** None. **C. de Oliveira:** None. **E. wong:** None. **S. Schmid:** None. **M. Hebb:** None.

Poster

395. Traumatic Brain Injury: Therapeutic Interventions II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 395.23/DD23

Topic: C.09. Brain Injury and Trauma

Support: FDA Office of Chief Scientist

FDA Critical Path

Title: Long-term viability of optogenetically transfected neurons and implantable electrodes in the motor cortex of mice

Authors: *C. GORINI[1], B. KOO[1], C. ALTIMUS[1], E. F. CIVILLICO[2]
[1]Food and Drug Administration/CDRH/OSEL/DBP, Silver Spring, MD; [2]OD/OSC, NIH, Rockville, MD

Abstract: Optogenetics has become a critical part of the neural circuit research toolbox; however, its utility for chronic studies, which is directly related to its therapeutic promise, has yet to be proven. Few studies have utilized optogenetics for chronic applications, and there is a paucity of knowledge regarding the stability of opsin expression or the neuronal network response to repeated stimulation over time. In order to contribute to the understanding of the long-term viability of optogenetic gene therapy in combination with implanted intracortical

devices, we performed bilateral injections of an adeno-associated virus (AAV5:hSyn:ChR2:EYFP) to transfect the excitatory opsin, channelrhodopsin-2(ChR2), into the mouse motor cortex. We then unilaterally implanted a 16 channel shank recording optrode (A16-177-OCM16, NeuroNexus) into one of the previously injected cortical hemispheres of each mouse. Following recovery from surgery, spontaneous cortical activity was recorded in freely moving mice. During weekly recording sessions, the cortex was illuminated with pulsed 473nm laser light (Laserglow Technologies) for 10ms, 20ms, 50ms, and 100ms, at 1Hz to excite ChR2+ motor neurons. Responses to light pulses were quantified to determine changes in neuron spike amplitude or firing frequency over time. After collection of data for 8-12 months, animals were sacrificed and tissue analyzed for changes in microglia response, neuroinflammation, and morphology. Viral injection sites with and without devices were compared to examine the interaction between the viral transfection and the foreign body response to the implant. Preliminary results do not indicate any significant changes in morphology or immune response when compared to the contralateral, unactivated hemisphere. In ongoing analysis, we are characterizing the dynamics of the cortical response to this multimodal intervention. **Disclaimer**: The mention of commercial products, their sources, or their use in connection with material reported herein is not to be construed as either an actual or implied endorsement of such products by the Department of Health and Human Services.

Disclosures: C. Gorini: None. **B. Koo:** None. **C. Altimus:** None. **E.F. Civillico:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.01/DD24

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Title: Safety of intrathecally administered baclofen in rats

Authors: *T. TAKENAMI[1], Y. NARA[2], R. ISONAKA[3], K. SUGIMURA[2], Y. SUZUKI[2], H. OKAMOTO[2]
[1]Kitasato Univ., Sagamihara, Japan; [3]Dept. of PhysiologyDepartment of Physiol., [2]Kitasato Univ. Sch. of Med., Sagamihara, Japan

Abstract: Background: Intrathecal infusion of baclofen (ITB) therapy for spasticity was approved application of insuarance in Japan at 2006. Recently, ITB is reported to be analgesic effect for chronic pain [1], central pain [2], or CRPS [3] regardless of spasticity. However, there is only little information on baclofen-related neurotoxicity. Therefore, we examined intrathecal baclofen neurotoxicity by histological and neurofunctional analyses. Methods: Thirty-three rats were each randomly injected intrathecally with 0.12μl/g body weight of either of 400 (400B), 800 (800B), 2000 (2000B), 3000 (3000B), 4000 (4000B), or 8000 (8000B) μg/ml of baclofen dissolved in

saline. Control rats received saline alone. Seven days after the injection, the L2 spinal cord with the anterior and posterior roots, dorsal ganglion, and cauda equina were extracted for microscopy. The hind limbs were evaluated neurologically by walking behavior. Results: Neurohistopathological examination showed no evidence of neurotoxic damage in all groups. Immediately after intrathecal baclofen injection, all rats displayed lower limbs weakness, and couldn't keep normal posture. However, all rats of each group could walk normally within 0.25, 0.25, 1, 1, 4, and 4h after injection in 400B, 800B, 2000B, 3000B, 4000B, and 8000B groups, respectively. None of the rats showed irreversible palsy or required tracheal intubation, even at 8000B. The sensory threshold was no significant difference among the baclofen and control groups at 7 days post-injection. Discussion and Conclusion: Intrathecal baclofen produced no histological or neurofunctional neurotoxicity even at 8000B. Our previous studies on 4% bupivacaine neurotoxicity using the same experimental protocol showed nerve damage in rats, and posterior nerve lesions at concentrations approximately 8 times higher than the clinically-used concentration [4], while 8000B is approximately 50 times higher concentration than clinical concentration. These results indicate that intrathecal baclofen is a safe treatment option for chronic intractable pain. However, further investigation is necessary to determine the effects of continuous long-term infusion of baclofen. References:1.RAPM 2004;29:269 2.Anesthesiology 2000;92:876 3.RAPM 2002;27:90 4.RAPM 2005;30:464

Disclosures: T. Takenami: None. **Y. Nara:** None. **R. Isonaka:** None. **K. Sugimura:** None. **Y. Suzuki:** None. **H. Okamoto:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.02/DD25

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: FAPESP - 2014/06892-3

CNPq - 300552/2013-9

Title: Anti-apoptotic effects of a fluorinated cannabidiol derivate: neuronal preservation and gliosis attenuation following neonatal sciatic nerve axotomy

Authors: S. A. OLIVEIRA[1], M. PEREZ[2], G. CHIAROTTO[1], R. MECHOULAM[3], F. S. GUIMARAES[4], *A. L. OLIVEIRA[5]
[1]Univ. of Campinas, Campinas, Brazil; [2]Unicamp, Campinas, Brazil; [3]Inst. for Drug Research, Med. Faculty, Hebrew Univ., Jerusalem, Israel; [4]Sch. Med. Ribeirao Preto-Usp, Ribeirao Preto, Brazil; [5]Univ. of Campinas - Lab. of Nerve Regeneration, Campinas, Brazil

Abstract: Lesion to the immature peripheral nervous system, such as the transection of a peripheral nerve, results in extensive degeneration of motoneurons and dorsal root ganglia (DRG) sensory neurons, mostly by apoptotic events. In turn, the axotomy of the sciatic nerve in the neonate has become a golden standard experimental approach to test new neuroprotective molecules and therapies. We have previously shown that cannabidiol (CBD), the most abundant non-psychotropic molecule present in Cannabis sativa leaves, exhibits neuroprotective action, reducing spinal motoneuron loss by 20% and dorsal root ganglia degeneration by 10% when daily administered at a dose of 15mg/Kg. The present work shows that the use of 4'-fluoro-cannabidiol, HUF-101, a fluorinated synthetic version of CBD, significantly improves neuronal survival by 2 to 3-fold with only one-third of the dose. Furthermore, we show that HUF-101 administration significantly upregulates anti-apoptotic genes and block pro-apoptotic nuclear factors expression. Thus, 2-day-old Wistar rats were subjected to a unilateral section of the sciatic nerve and daily treated with HUF-101 (1, 2.5, 5 mg/Kg/day, i.p.) or vehicle solution for five days. The results were evaluated by Nissl staining, immunohistochemistry, and qRT-PCR. Neuronal counting revealed 40 % rescue of spinal motoneurons and 20% improved preservation of DRG neurons (HUF-101, 5mg/Kg) when compared to the vehicle treatment. Such survival was associated with complete depletion of p53 and an elevation of 60-fold of BCL2 like 1 gene expression. Also, peroxisome proliferator-activated receptor gamma (PPAR-gamma) gene expression was downregulated by 80%. Neuronal preservation was coupled with a reduction in astroglial reaction, studied by glial fibrillary acidic protein (GFAP) immunolabeling, which shown a 50% reduction when the integrated density of pixels was evaluated nearby spinal motoneurons, present in the ventral horn of the lumbar intumescence. Overall, the current data strongly indicates the HUF-101 has potent neuroprotective effects, significantly superior to native CBD. Such properties are particularly related to anti-apoptotic protection and reduction of astrogliosis.

Disclosures: **S.A. Oliveira:** None. **M. Perez:** None. **G. Chiarotto:** None. **R. Mechoulam:** None. **F.S. Guimaraes:** None. **A.L. Oliveira:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.03/DD26

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant CA1832736

NIH Grant NS073939

NIH Grant CA20871

Acetylon Pharmaceuticals Inc.

Title: Inhibition of HDAC6 reverses cisplatin-induced neurotoxicity

Authors: *J. MA[1], K. KRUKOWSKI[1], O. GOLONZHKA[2], G. LAUMET[1], T. GUTTI[1], J. VAN DUZER[2], R. MAZITSCHEK[3], M. JARPE[2], C. J. HEIJNEN[1], A. KAVELAARS[1]
[1]Symptom Res., MD Anderson Cancer Ctr., Houston, TX; [2]Acetylon Pharmaceuticals Inc, Boston, MA; [3]Massachusetts Gen. Hosp., Boston, MA

Abstract: Chemotherapy-induced neurotoxicity is among the most common dose-limiting side-effects of cancer treatment, which not only hampers effective cancer therapy, but also greatly reduces the quality of life for cancer survivors. Currently, there is no FDA-approved treatment available. Histone deacetylase 6 (HDAC6) is a microtubule-associated deacetylase whose function includes regulation of α-tubulin-dependent intracellular mitochondrial transport. Inhibition of HDAC6 has been shown protective in several neurological disorders. Here we examined the effect of HDAC6 inhibition on established cisplatin-induced peripheral neuropathy and cisplatin-induced cognitive impairments in mice. We used a novel HDAC6 inhibitor ACY-1083, which readily crosses the blood-brain barrier and shows 260-fold selectivity towards HDAC6 versus other HDACs. The HDAC6 inhibitor was injected intraperitoneally for 2 weeks after termination of cisplatin treatment. Our results demonstrate that HDAC6 inhibition completely reversed already existing cisplatin-induced mechanical allodynia, spontaneous pain, numbness, and cognitive deficits. It also restored the loss of intra-epidermal nerve fiber density in cisplatin-treated mice. Mechanistically, treatment with the HDAC6 inhibitor increased α-tubulin acetylation. In addition, HDAC6 inhibition restored the cisplatin-induced reduction in mitochondrial bioenergetics and mitochondrial content in the tibial nerve, indicating increased mitochondrial transport. At a later time point, dorsal root ganglion mitochondrial bioenergetics also improved. Cisplatin-induced changes in mitochondrial morphology and bioenergetics in the brain synaptosomes were also restored by the HDAC6 inhibitor. Our results demonstrate that pharmacological inhibition of HDAC6 completely reverses established cisplatin-induced peripheral neuropathy and cognitive impairments, and the protective effects were associated with normalization of mitochondrial function. These results are especially promising because HDAC6 inhibitors are currently used in clinical trials as an add-on cancer therapy, highlighting the potential for clinical translation of our findings.

Disclosures: **J. Ma:** None. **K. Krukowski:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent. **O. Golonzhka:** A. Employment/Salary (full or part-time):; Acetylon Pharmaceuticals. **G. Laumet:** None. **T. Gutti:** None. **J. van Duzer:** A. Employment/Salary (full or part-time):; Acetylon Pharmaceuticals. **R. Mazitschek:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Acetylon Pharmaceuticals. **M. Jarpe:** A. Employment/Salary (full or part-time):; Acetylon Pharmaceuticals. D. Fees for Non-CME Services Received Directly from Commercial Interest or their Agents (e.g., speakers' bureaus); patent holder. **C.J. Heijnen:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship

even if those funds come to an institution.; Grant from Acetylon Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent holder. **A. Kavelaars:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Grant from Acetylon Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent holder.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.04/DD27

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: Koshiyama Research Grant

Title: Neuroprotective effects of novel oxyindole compounds against oxytosis, feroptosis and ER stress

Authors: *Y. HIRATA[1], C. YAMADA[1], Y. ITO[1], S. YAMAMOTO[1], H. NAGASE[1], K. OH-HASHI[1], K. KIUCHI[1], H. SUZUKI[2], M. SAWADA[2], K. FURUTA[1]
[1]Gifu Univ., Gifu, Japan; [2]Nagoya Univ., Nagoya, Japan

Abstract: Current medical and surgical therapies for neurodegenerative diseases such as Alzheimer's disease and Parkinson's disease offer symptomatic relief but do not provide a cure. Small synthetic compounds that protect neuronal cells from degeneration are critically needed to prevent and treat the aforementioned diseases. Both oxidative stress and endoplasmic reticulum (ER) stress have been implicated in the pathophysiological conditions of those neurodegenerative diseases. In search of neuroprotective agents against oxidative stress and/or ER stress using the murine hippocampal HT22 cell line, we found a novel oxindole compound, GIF-0726-r. The structure is comprised of an indane-type skeleton and an electron-rich unsaturated side-chain attached at C-1 of the ring system. It is effective in preventing limited types of cell death involved in neurodegenerative disorders, such as Alzheimer's and Parkinson's diseases. GIF-0726-r prevented oxidative stress, including glutamate-induced oxytosis and erastin-induced ferroptosis, and tunicamycin-induced ER stress, but did not affect apoptosis induced by staurosporine, camptothecin, and etoposide. GIF-0726-r was capable of inhibiting reactive oxygen species generation and Ca^{2+} influx, a presumed executor in cell death, and activating the antioxidant response element (ARE), a *cis*-acting regulatory element in promoter regions of several genes encoding phase II detoxification enzymes and antioxidant proteins. In addition, GIF-0726-r reduced ER stress-induced gene expression of GADD153 and Grp78 and

XBP1 mRNA splicing for the key downstream targets of ER stress sensors. These results suggest that GIF-0726-r is a low molecular weight compound that prevents neuronal cell death through the attenuation of oxidative and ER stress pathways, and could be a useful therapeutic, as well as a pharmacological tool to manipulate the cellular stress response. We prepared over two hundred oxindoles and related compounds by the modification of GIF-0726-r structure and screened those according to the ARE activity. GIF-2651X-G1 was the most potent ARE activator and successfully showed neuroprotective activity *in vivo* in a model of Parkinson's disease. Further studies are needed to assess the efficacy of GIF-2651X-G1 using various animal models of Parkinson's and Alzheimer's diseases.

Disclosures: **Y. Hirata:** None. **C. Yamada:** None. **Y. Ito:** None. **S. Yamamoto:** None. **H. Nagase:** None. **K. Oh-hashi:** None. **K. Kiuchi:** None. **H. Suzuki:** None. **M. Sawada:** None. **K. Furuta:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.05/DD28

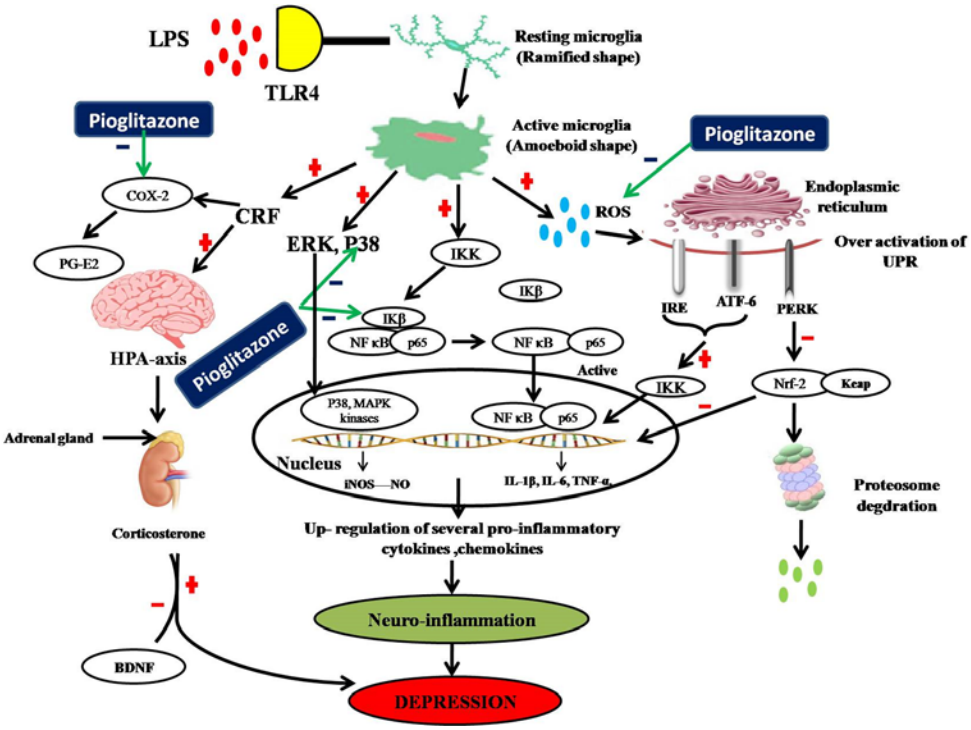
Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Title: Pioglitazone attenuates lipopolysaccharide- induced neuroinflammation and depressive-like behaviour via suppressing the NFκB/Nrf-2 and MAPK signalling pathways

Authors: *S. AHMED[1], M. KWATRA[1], A. AHMED[2], V. NAIDU[1], M. LAHKAR[1], B. BEZBARUAH[3]
[1]Pharmacol. and toxicology, NIPER, Guwahati, India; [2]Pathology, Rajarajeshwari Med. Col., Bangalore, India; [3]Gauhati Med. Col., Guwahati, India

Abstract: Depression is generally described as a low mood state and repugnance to physical activity provoking for suicidal tendencies. According to WHO report, nearly 450 million people suffer from mental disorders of which nearly 10-20 million commit suicide every year. Despite having several medications the disease is still a challenging with approximately a large number of populations do not respond to their first line medication. Hence there exists a necessity to explore new targeted drugs. Lipopolysaccharide (LPS) is an endotoxin obtain from gram negative bacteria which, when injected into experimental mice produces depressive-like behaviour by activating the microglia cells. Upon activation, microglia releases a plethora of mediators including cytokines, chemokines, reactive oxygen species (ROS) and reactive nitrogen species (RNS) which may alter the serotoninergic and glutamatergic neurotransmission. Pioglitazone belongs to peroxisome proliferator-activated receptor gamma (PPAR-γ) agonist class regulates lipid metabolism, exerts potent central and peripheral anti-neuroinflammatory action and possesses neuroprotective effect. However, the mechanisms through which PPAR's

exert these effects have yet to be fully elucidated. In this study, we have explored the neuroprotective nature of pioglitazone in attenuating these insults via inhibiting the activation of multiple transcription factors such as NF-κB, Nrf-2 and MAPKs. Pioglitazone at a dose of 30mg/kg body weight for 14 days was sufficient to show neuroprotective activity by inhibiting the oxido-nitrosative stress and other inflammatory cytokine markers. Taken together our study concludes the neuroprotective potential of pioglitazone in psychiatric disorders associated with inflammation and oxidative stress.



Disclosures: **S. Ahmed:** None. **M. Kwatra:** None. **A. Ahmed:** None. **V. Naidu:** None. **M. Lahkar:** None. **B. Bezbaruah:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.06/DD29

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: RDA Grant PJ01100201

Title: Restoring the impaired cholesterol accumulation and autophagy by graphene enhances neuronal differentiation and prolongs survival in Niemann pick type C disease

Authors: *K.-S. KANG
Vet. medicine, Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: Niemann-Pick type C (NPC) disease is an inherited lipid storage disorder, characterized by neurodegeneration. Mutation in NPC1 causes accumulation of unesterified cholesterol and dysregulation of autophagy. Here, we report NPC1 mutant mice given graphene show improved clinical outcomes through improved neuronal survival, especially Purkinje cells. In survived Purkinje neurons, cholesterol accumulation was significantly decreased. Interestingly, this therapeutic effect was also found in generated human induced neural stem cells from NPC patient-derived fibroblasts (NPC-iNSC). NPC-iNSC treated with graphene showed restored cholesterol accumulation and improved neuronal differentiation. Additionally, we were able to observe that dysfunctional autophagic flux in NPC-iNSC was restored by graphene. Taken together, graphene can rescue of homeostatic regulation of cholesterol storage and autophagy in NPC disease and provide a potential therapeutic treatment option for neurodegenerative diseases.

Disclosures: K. Kang: None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.07/DD30

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: Howard Hughes Medical Institute

Title: Insulin containing PLGA nanoparticles may restore myelin to axon area and proprioceptor function in diabetic rat

Authors: *V. K. HAFTEL, C. BOWE, A. NWANCHA, A. OLADIMEJI, R. MCREYNOLDS, T. GYLES
Dept of Biol., Morehouse Col., Atlanta, GA

Abstract: Diabetic neuropathy is a complication that can arise from diabetes that commonly causes nerve degeneration in the peripheral regions of the body. As a result, a person can experience a number of symptoms such as pain, numbness, a loss of reflexes and a general loss of sensation, which can result in serious infections. It has been suggested that insulin can bind to receptors on the nerves to encourage growth, therefore, may be possible to prevent or counteract this degeneration by delivering insulin directly to the degenerating nerves. One possible method of delivering this insulin to the nerves is through the use of nanoparticles (InsNP). Using the biodegradable NPs will allow the insulin to be primarily deposited at the site of interest with

little to no consequence. In our experiments, the effects of diabetic neuropathy were observed using the ratio of the area of the myelin sheath to the area of the axon (m:a), along with electrophysiological measures of individual proprioceptor responses to muscle stretch from sciatic nerve of rats of various treatment groups. Data from untreated, short-term diabetic rats (3wk, 6wk) and diabetic rats with InsNP injected in triceps surae muscles was reanalyzed and sham-injected control groups were added. It has been observed that diabetes does have an effect on the m:a and that nanoparticle injections of insulin bring the ratio towards the non-diabetic control ratios. In order to ensure that the effects were a result of the InsNP injections, and not the injection into muscle, itself, a control group was added: diabetic rats with triceps surae injections of phosphate buffer solution (PBS). These controls are tested because it is possible that the perceived recovery of the m:a and electrophysiological measurements following InsNP injection may be a result of a reaction to the injection or the PBS in which NPs are suspended. In fact, the data show that m:a was much reduced in the 3wk diabetic + sham injected rats (3.6 +/- 0.05 vs. 5.3 +/- 0.12 for 3wk diabetic, $p = 0.00002$; 8.9 +/- 0.07 untreated, $p = 0.00002$; 5.7+/- 0.09 for 3wk diabetic + 1 wk InsNP, $p = 0.89$, ANOVA, post hoc unequal N HSD), therefore, not explaining the recovery toward untreated values of m:a in the InsNP injected rats. In future experiments we aim to examine the time course of InsNP treatment more closely, test various formulations of NPs to possibly increase the stability of insulin release, and to determine whether or not insulin growth factor (IGF1) is more effective than insulin in the prevention or treatment of diabetic neuropathy.

Disclosures: V.K. Haftel: None. **C. Bowe:** None. **A. Nwancha:** None. **A. Oladimeji:** None. **R. McReynolds:** None. **T. Gyles:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.08/DD31

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: NASA Grants NNX13AB73G and NNX16AE06G

Title: Exposure to low doses of helium particles disrupts neuronal function and cognitive performance

Authors: *B. M. RABIN[1], S. M. POULOSE[2], M. G. MILLER[2], D. F. BIELINSKI[2], K. L. CARRIHILL-KNOLL[1], E. M. HAWKINS[1], R. C. HENG[1], A. LARSEN[1], C. SPADAFORA[1], N. N. ZOLNEROWICH[1], R. PATEL[1], B. SHUKITT-HALE[2]
[1]Univ. Maryland Baltimore County, Baltimore, MD; [2]Neurosci., HNRCA At Tufts Univ., Boston, MA

Abstract: On exploratory class missions, such as a mission to Mars, astronauts will be exposed to types of radiation (cosmic rays) that are not experienced in low earth orbit where the space shuttle and International Space Station operate. A significant portion of this radiation will be composed of low linear energy transfer (LET) helium (^{4}He) particles, which produce less damage to neuronal tissue as they pass through the brain. Sprague-Dawley rats weighing between 200-225 g were given either head-only or whole-body exposures to ^{4}He particles (1000 MeV/n) at doses ranging from 0.01 to 0.5 cGy. Following exposure, the brains were removed from a subset of the rats for analysis of oxidative stress by measuring NADPH oxidase (NOX2) expression. Measurements were also made of phosphorylated-cyclic AMP (cAMP)-responsive element-binding protein (CREB) and nuclear factor E2-related factor 2 (Nrf2). The remaining rats (n = 10/dose) were shipped to UMBC for behavioral testing, including elevated plus maze (baseline anxiety); novel object and novel spatial recognition (learning and memory); and operant responding on an ascending fixed-ratio schedule (motivation to work for reward). The results indicated that changes in behavioral endpoints could be observed following exposures to ^{4}He particles at doses as low as 0.01 to 0.025 cGy. Differences in neurochemical endpoints were observed following exposure to doses as low as 0.05 cGy (the lowest dose tested). There were no significant differences between head-only and whole-body exposures on behavioral performance. Preliminary analyses of the neurochemical data suggest that whole body exposures may not have been as effective in producing changes in neuronal functioning as were head-only exposures. Because ^{4}He particles will constitute a significant fraction of the radiation dose to which astronauts will be exposed, the present results suggest the possibility that astronauts on exploratory class missions may be at a greater risk for HZE-induced cognitive deficits than anticipated.

Disclosures: B.M. Rabin: None. **S.M. Poulose:** None. **M.G. Miller:** None. **D.F. Bielinski:** None. **K.L. Carrihill-Knoll:** None. **E.M. Hawkins:** None. **R.C. Heng:** None. **A. Larsen:** None. **C. Spadafora:** None. **N.N. Zolnerowich:** None. **R. Patel:** None. **B. Shukitt-Hale:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.09/DD32

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: HRF-201701-017

HRF-S-53

Title: Long-term effects of NADPH oxidase inhibitor treatment on seizure-induced hippocampal neurogenesis

Authors: *S. LEE[1], B. CHOI[1], S. SUH[1], M. LEE[2], H. SONG[2], H. CHOI[2], M. SOHN[3]
[1]Dept. of Physiol., [2]Dept. of Neurol., Hallym Univ., Chuncheon, Korea, Republic of; [3]Dept. of Nursing, Inha Univ., Incheon, Korea, Republic of

Abstract: Apocynin, known as acetovanillone, is a natural organic compound structurally related to vanillin. Apocynin also known as an inhibitor of NADPH oxidase activity and thus is effective in preventing the production of superoxide. Apocynin has been studied to determine its application in several brain injuries, such as stroke, traumatic brain injury (TBI) and epilepsy. Previously, our lab demonstrated that TBI or seizure-induced oxidative injury and neuronal death were reduced by apocynin treatment. Several studies have demonstrated that neuroblast production is increased in the hippocampus after seizure. Here we confirmed the hypothesis that long-term treatment with apocynin may enhance newly generated hippocampal neuron survival by reduction of superoxide production after seizure. Seizure was induced by pilocarpine (25mg/kg i.p.) injection. Apocynin was continuously injected for 4 weeks after seizure (once per day) into the intra intraperitoneal space (i.p.). We evaluated NeuN, BrdU and doublecortin (DCX) immunostaining to determine whether treatment of apocynin increased neuronal survival and neurogenesis in the hippocampus after seizure. The present study indicates that long-term treatment of apocynin increased the number of NeuN (+) and DCX (+) cells in the hippocampus after seizure. Therefore this study suggests that apocynin treatment increased neuronal survival and neuroblast production by reduction of hippocampal oxidative injury after seizure.
Keywords: Epilepsy, pilocarpine, neuron death, NADPH-oxidase inhibitor, oxidative stress, neurogenesis

Disclosures: S. Lee: None. **B. Choi:** None. **S. Suh:** None. **M. Lee:** None. **H. Song:** None. **H. Choi:** None. **M. Sohn:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.10/DD33

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: HRF-201701-017

HRF-S-53

Title: Effects of donepezil, an acetylcholinesterase inhibitor, in hippocampal neurogenesis after pilocarpine-induced seizure

Authors: *J. JEONG[1], B. CHOI[1], S. SUH[1], H. CHOI[2], H. SONG[2]
[1]Dept. of Physiol., [2]Dept. of Neurol., Hallym Univ., Chuncheon, Korea, Republic of

Abstract: Epileptic seizures are short episodes of abnormal brain electrical activity and the presence of recurrent and unprovoked seizures is known as epilepsy. Primary symptoms of epileptic seizure may include tonic-clonic movement or convulsions, accompanied by a loss of memory that may be short or sustained. Many survivors of severe epilepsy display delayed neuronal death and permanent cognitive impairment. It is well known that seizure increases the rate of adult neurogenesis in the subgranular zone of the dentate gyrus of the hippocampus. Severe seizures rapidly induce an increase in the proliferation of neural progenitor cells. However, the exact mechanism by which seizures regulate progenitor cell proliferation and neurogenesis is not well known. Donepezil, also called Aricept, is an acetylcholinesterase inhibitor. Donepezil is an effective treatment agent for Alzheimer's disease, dementia and in ischemic or hypoxic settings. However, the role of donepezil on seizure-induced neurogenesis remains untested. Thus, the present study sought to evaluate the therapeutic potential of donepezil treatment on seizure-induced neural injury. Temporal lobe epilepsy (TLE) was induced by intraperitoneal (*i.p*) injection of pilocarpine (25 mg/kg) in male rats. Behavioral testing confirmed the presence of seizure. Donepezil (2.5 mg/kg/day) was injected by gavage for one week after seizure to see the effect of short-term administration. In addition, donepezil was injected for 3 weeks from 3 weeks after seizure to observe the effects of long-term administration. One or six weeks after seizure, BrdU staining and DCX staining were performed to evaluate the effect of donepezil on neurogenesis. There was no difference in the number of BrdU and DCX positive cells when donepezil was administered for one week after seizure. However, the number of BrdU and DCX positive cells was increased in the donepezil-treated group, compared to the vehicle-injected group, when evaluated at six weeks after seizure. In conclusion, donepezil was not effective in the short time period after seizure but was effective in promoting neurogenesis when administered for longer timeframes after seizure. Keywords: Epilepsy, pilocarpine, neurogenesis, donepezil

Disclosures: **J. Jeong:** None. **B. Choi:** None. **S. Suh:** None. **H. Choi:** None. **H. Song:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.11/DD34

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Title: Acute and sustained excitoxicity differentially influence riluzole’s neuroprotective effect

Authors: *S. WAGNER, E. ANDRIAMBELOSON, C. NEVEU
NEUROFIT, ILLKIRCH, France

Abstract: Excitotoxicity is caused by overstimulation of glutamate receptors. This pathological process that results from excessive release glutamate leads to neuronal injury or death in various

neurological disease conditions. An acute elevation of glutamate is thought to induce neuronal injury in conditions such as stroke, epilepsy and brain trauma injury. More chronic and sustained exposure to milder elevations of glutamate is believed to mediate excitotoxicity in neurodegenerative diseases such as Amyotrophic lateral sclerosis (ALS).The aim of the present study was to compare the excitotoxic effect of acute (10 - 60 min) and sustained (24 h) exposure to glutamate by cultures of cortical neurons. The neuroprotective effect of riluzole (the unique FDA-approved drug ALS drug) was comparatively assessed under the two glutamate exposure conditions. Two biochemical measurements were used to evaluate glutamate-induced neuronal damage 1 day after the initiation of excitotoxicity: *i)* measure of the increase in the amount of lactate dehydrogenase (LDH) released in the cell culture supernatant and *ii)* measure of the decrease in the ATP level in the cell culture.Acute exposure of cortical neurons to glutamate (3, 10, 20 and 75 µM) produced a concentration-dependent increase in extracellular LDH (up to 2-fold increase as compared to the control). When the exposure was extended to 24 h, the amount of extracellular LDH became even more elevated (up to 4-fold increase). Whilst riluzole treatment did not prevent the increase in LDH observed after acute glutamate exposure, it fully abolished the further increase in LDH induced by the sustained exposure. These results suggest that sustained exposure to glutamate induces an additional mechanism of death which is sensitive to riluzole, unlike the one caused by acute exposure. A similar pattern of results was observed when using the decrease of ATP level as a measure of neuronal damage.The above results suggest that acute and sustained exposure to glutamate induce differential mechanisms of neuronal death and this finding is supportive of the beneficial effect of riluzole in neurodegenerative diseases such as amyotrophic lateral sclerosis where more chronic exposure to glutamate is hypothesized.

Disclosures: S. Wagner: None. **E. Andriambeloson:** None. **C. Neveu:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.12/DD35

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: National Research Foundation (NRF) of Korea (NRF-2017R1A2B3005753)

Title: Neuroprotective effects of lipid emulsion in the acute phase of kainic acid-induced injury in the rat hippocampus

Authors: *M. TANIOKA[1,2], S. UM[1,2], K. KIM[1,2], B. LEE[1,2]
[1]Yonsei University, Grad. Sch. Dept. of Med. Sci., Seoul, Korea, Republic of; [2]Brain Korea 21 PLUS Project for Med. Sci., Yonsei University, Seoul, Korea, Republic of

Abstract: Numerous anti-epileptic drugs (AEDs) have been successful in attenuating seizures. However, neuroprotective methods for the neural injuries that occur after seizures are still in jeopardy due to the lack of specificity in targeting protective markers. Excitotoxic substances such as kainic-acid (KA) and pilocarpine are widely used to induce status epileptus (SE) in animal models causing permanent impairments in the brain. In recent research, lipid emulsion (LE), a well-known parental nutrition has attenuated excitotoxic cell death caused by local anesthetics in myocardial cells. Intravenous LE was reported to provide cytoprotection via various mechanisms involving fatty acid interactions at molecular levels. In our study, we tested the cytoprotective effect of LE on excitotoxic injury in the brain and investigated its protective mechanisms. This study used male Sprague Dawley rats which were divided into 4 groups: vehicle, KA+vehicle, KA+LE 0.01%, and KA+LE 1%. The experimental animals underwent guide cannulae implantation surgeries under pentobarbital anesthesia. Bilateral intrahippocampal injections of KA and LE were given to the experimental groups. Protective effects were observed for both LE-pretreated animals, and animals that were treated with LE after the induction of acute seizures. After the treatment of the experimental groups, the elevated plus maze and passive avoidance behavioral tests were performed and analyzed. After the behavioral tests, brains were perfused and stained with cresyl violet to measure morphological changes. As a result, LE with the concentration of 1% increased the survival of neurons in the pyramidal tract of the hippocampus. Behavioral analysis also displayed less impairment of cognition in the passive avoidance test and decreased the level of anxiety in the elevated plus maze compared to the control group. Wnt1 was up-regulated as a potent protective biomarker at molecular levels which may be due to the palmitate bonding provided by LE. As a result, the canonical Wnt signaling pathway can be a potential target for neuroprotection via fatty acid interaction. Our data suggest that the nutritional aspects of phytochemicals may provide neuroprotection. Further studies that accompany phytochemicals with existing AEDs may lead to new treatments and possibly aid patients suffering from cognitive deficits caused by neurodegenerative diseases.
This work was supported by the National Research Foundation (NRF) of Korea funded by the Ministry of Science, ICT, and Future Planning (NRF-2017R1A2B3005753).

Disclosures: **M. Tanioka:** None. **S. Um:** None. **K. Kim:** None. **B. Lee:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.13/DD36

Topic: C.01. Brain Wellness and Aging

Title: Pharmacological agents for enhancing cognitive functions

Authors: *G. B. PATRUDU
Andhra Med. Col. & King George Hosp., Visakhapatnam, India

Abstract: Research on the use of drugs for cognitive enhancement has been on the rise. Several classes of drugs can positively influence cognitive functions. This review classifies the various classes of drugs that have been used in cognitive enhancement research based on their mechanism of action.The classes of drugs that have been used in studies include Psychostimulants (amphetamine, methylphenidate, modafinil, caffiene, nicotine etc); Drugs increasing availability of various neurotransmitters in the brain like Acetylcholinesterase inhibitors (donepezil, galantamine, rivastigmine), sources of choline (citicholine), noradrenaline reuptake inhibitors (reboxetine and atomoxetine), MAO inhibitors, Tricyclic antidepressants, serotonin noradrenaline reuptake inhibitors (duloxetine) ,Selective seratonin reuptake inhibitors (Fluoxetine),Central dopamine increasing agents (levodopa); central vasodilators (Nimodepine); drugs acting on Glutamate receptors (riluzole); NMDA partial antagonists (memantine); Drugs acting on AMPA receptor (Ampkines, Racetams); Drugs increasing cAMP in the brain such as PDE4 inhibitors(roflumilast); drugs enhancing BDNF production such as Histone deactylase (HDAC) inhibitors (valproate); drugs enhancing metabolism in the brain mitochondria (Methylene blue); Growth factors involved in neurogenesis (Insulin like growth factor (IGF), Brain derived Neurotrophic factor(BDNF) etc); Drugs activating CREB (Lithium) and opiate receptor antagonists(naloxone, substance P).

Disclosures: G.B. Patrudu: None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.14/EE1

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Title: A clinically relevant, low-dose activated protein C (APC) protects the brain against ischemic stroke in mice

Authors: *K. YAMATO[1], Y. NAKAJO[1,4], H. YAMAMOTO-IMOTO[2], K. KOKAME[2], T. MIYATA[2], H. KATAOKA[3], J. C. TAKAHASHI[3], H. YANAMOTO[1,5]
[1]Lab. of Neurol. and Neurosurg., [2]Dept. of Mol. Pathogenesis, [3]Dept. of Neurosurg., Natl. Cerebral and Cardiovasc. Ctr., Suita, Japan; [4]Res. Lab., Rakuwa–kai Otowa Hosp., Kyoto, Japan; [5]Dept. of Cardiovasc. Science, Div. of Surgical Med., Osaka Univ. Grad. Sch. of Med., Suita, Japan

Abstract: Plasma levels of protein C (PC) are inversely associated with the incidence of ischemic stroke, according to a large prospective study, suggesting that the serine protease:

activated PC (APC), an intrinsic anticoagulant, may have a role in protecting the brain against ischemic stroke. Previously, we demonstrated that 2 mg/kg APC (i.v.) in the acute phase, rather than higher doses, suppresses the development of cerebral infarction after temporary focal ischemia, with some cases of hemorrhagic transformation. Here, we examined the effect of < 2 mg/kg APC, using the same three-vessel occlusion (3-VO) model (Yang *et al.*, 2014). The effect of the potent anticoagulant, heparin, or a protease-activated receptor (PAR)-1 antagonist, SCH79797, was also investigated in this model. Male C57BL/6J mice, received either human APC (Kaketsuken) - 0.25, 0.5, 1 or 2 mg/kg, heparin - 600 U/kg, or saline (i.v.), at 5 min and 3 h (twice for the acute phase analysis), or at 5 min, 3, 24, 48, and 72 h (five times for the chronic phase analysis), after the induction of ischemia. The regional cerebral blood flow (rCBF) was monitored using laser Doppler flowmetry. Infarcted lesion sizes and neurological deficits were analyzed at day 1 or 7. The suppression in the infarcted lesion sizes was most prominent in the 0.5 mg/kg APC group, in which the rCBF during ischemia was not affected, neurological deficits were less severe compared to the control, and no hemorrhagic transformation occurred. The treatment with heparin did not suppress the lesion volume, and the pretreatment with SCH79797 at 1 mg/kg (i.p.), 1 h before the induction of ischemia, abolished the efficacy of the 0.5 mg/kg APC. It has been reported that 2 mg/kg APC, or 600 U/kg heparin, but not < 2 mg/kg APC, was effective in suppressing the development of cerebral infarction in the intra-luminal thread-insertion (ITI) model used for assessing neuroprotective properties, indicating that APC or heparin prevents the thread (foreign body)-induced blood coagulation, prevents reduction in the rCBF and improves the outcome. However, the determined efficacy of heparin in the present study confirmed that the coagulation system does not play a role in inducing ischemia in our 3-VO model and that APC acts as a pure neuroprotectant, i.e., protects the brain without improving rCBF. As the pretreatment with the PAR-1 antagonist canceled the efficacy of APC, PAR-1-signaling is needed for the APC-derived neuroprotection. The clinically relevant low-dose APC, never demonstrated effective in the ITI model, was found to protect the brain against ischemic stroke, without causing hemorrhagic transformation.

Disclosures: **K. Yamato:** A. Employment/Salary (full or part-time):; Lab. of Neurol. and Neurosurg.,. **Y. Nakajo:** None. **H. Yamamoto-Imoto:** None. **K. Kokame:** None. **T. Miyata:** None. **H. Kataoka:** None. **J.C. Takahashi:** None. **H. Yanamoto:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.15/EE2

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Title: Hypoxia induces bidirectional plasticity in retinocollicular synapses

Authors: *H. DUMANSKA[1], N. VESELOVSKY[2]
[1]Dept. of Neuronal Networks Physiol., Bogomoletz Inst. of Physiology, Natl. Acade, Kyiv, Ukraine; [2]Dept. of Neuronal Networks Physiol., Bogomoletz Inst. of Physiology, Natl. Acad. of Sci. of Ukraine, Kyiv, Ukraine

Abstract: In this study we investigated effects of short-term hypoxia on synaptic transmission in the first visual (retinocollicular) synapses in coculture. Coculture was developed as adequate in vitro model of retinocollicular pathway with easily identified synaptic couples of retinal ganglion cell (RGC) - SC neuron. Method of fast local superfusion was used for short-term (5 minutes) application of hypoxic solution on synaptically connected neurons. Pharmacologically isolated evoked NMDA receptor (NMDAR)- and $GABA_AR$-mediated postsynaptic currents (PSCs) were and recorded from SC neurons by generation an action potential in presynaptic RGCs. Application of hypoxic magnesium-free solution resulted in a long-term potentiation (LTP) of NMDAR-mediated transmission. Furthermore, application hypoxic standard solution (2 mM Mg^{2+}) resulted in reduction in the ability of Mg^{2+} to induce a normal voltage-dependent blockade of the evoked NMDA response. Analysis of the oxygen deprivation effect on spontaneous and miniature postsynaptic currents (sPSCs and mPSCs) revealed an increase in occurrence frequency and the second peak appearance in the mPSCs histogram. Obtained estimates for quantal and binomial parameters reflected the presynaptic changes during the potentiation and can be caused by increasing in the average number of release sites and reducing of Mg^{2+} blockade. $GABA_AR$-mediated synaptic transmission responded to hypoxia by a long-term depression (LTD). Analysis of sPSCs and mPSCs showed significant decrease in the occurrence frequency and in the amplitude of mPSC (quantal size) during oxygen deficiency. Estimation changes in quantal and binomial parameters showed complex of presynaptic (independent of the release probability) and postsynaptic (decrease in sensitivity of postsynaptic receptors) mechanisms during hypoxia induced LTD. Physiological role of GABAergic retinocollicular projections is in regulation of activation and plasticity of excitatory NMDAR-mediated transmission. Thus, hypoxia induced LTD of GABAergic transmission could enhance pathological effect of LTP of NMDAR- mediated transmission in retinocollicular synapses and possibly is an additional injury which can be caused by hypoxia on transmission from the retina to subcortical visual center.

Disclosures: H. Dumanska: None. **N. veselovsky:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.16/EE3

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH T32HL125239

NIH R01NS060307

NIH K08NS080984

American Heart Association Grant-in-Aid

Title: Striatal neuronal death is attenuated in a swine model of pediatric hypoxic-asphyxic cardiac arrest with delayed hypothermia but off-target neurodegeneration is possible

Authors: *C. O'BRIEN[1], E. KULIKOWICZ[1], M. REYES[1], P. SANTOS[1], S. KANNAN[1], R. C. KOEHER[1], L. J. MARTIN[2], J. K. LEE[1]
[1]Anesthesiol. and Critical Care Med., [2]Pathology, Johns Hopkins Univ., Baltimore, MD

Abstract: Therapeutic hypothermia (HT) induced 4.9-5.9 h after resuscitation from cardiac arrest did not improve neurological outcomes in pediatric patients, in contrast to its benefit in perinatal hypoxia-ischemia. Whereas HT reduces necrotic cell death, it can promote apoptosis in cortical neurons and white matter after hypoxic-asphyxic (HA) cardiac arrest in piglets. Here, we tested the hypothesis that in putamen, a region vulnerable to necrotic cell death after HA, HT would decrease necrotic neuronal death but increase apoptotic cell death. We randomized 3-5 day old, male piglets to sham surgery or HA cardiac arrest followed by sustained overnight HT (34°C induced at 2 h), 20 h HT+8 h rewarming at 0.5°C/h, or normothermia. Viable, ischemic necrotic, and apoptotic profiles were counted in putamen. Sham normothermic (n=8) and naïve, unanesthetized (n=5) pigs had similar viable and ischemic neuron counts ($p>0.05$), but naïve pigs had more apoptosis than shams ($p=0.019$). Therefore, the anesthetic did not increase putamenal neuronal death. Sustained HT (n=7) and HT+rewarming (n=8) after HA preserved viable neuron counts to levels exceeding that of normothermia after HA (n=6; $p<0.001$ for both). Sustained HT and HT+rewarming after HA also reduced the number of ischemic necrotic neurons compared to normothermia ($p=0.007$ and $p=0.037$, respectively). After HA, the number of apoptotic profiles was similar among HT, HT+rewarming, and normothermic groups ($p>0.05$). However, among shams, sustained HT (n=7) increased apoptosis above that in normothermic (n=8) or HT+rewarmed (n=7) shams ($p=0.002$ for both). Interestingly, both sustained HT and HT+rewarming shams had decreased viable neuron counts relative to normothermic shams ($p<0.001$ and $p=0.019$, respectively). We conclude that HT with 2 h delayed induction protects putamenal neurons after HA cardiac arrest and, in contrast to cortical neurons, does not promote apoptosis. Insufficient neuroprotection from HT in clinical trials may be related to the 4.9-5.9 h delay in HT induction. Unexpectedly, HT independent of HA injury promoted putamenal apoptosis with loss of total viable neurons. Potential deleterious effects of HT in the developing brain warrants further study because cooling children with only mild hypoxic brain injuries may be detrimental.

Disclosures: C. O'Brien: None. **E. Kulikowicz:** None. **M. Reyes:** None. **P. Santos:** None. **S. Kannan:** None. **R.C. Koeher:** None. **L.J. Martin:** None. **J.K. Lee:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.17/EE4

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH K08NS080984

Title: Insufficient proteasome function: A possible cause of white matter injury during hypothermia for neonatal hypoxic-ischemic encephalopathy

Authors: *P. SANTOS
ACCM, Johns Hopkins Univ., Hanover, MD

Abstract: White matter injury occurs in neonatal encephalopathy despite the use of therapeutic hypothermia (1). White matter apoptosis is also triggered by whole-body mild hypothermia and rewarming after global hypoxia-ischemia (HI) in our neonatal piglet model of HIE (2). Because HI may interrupt the unfolded protein response (UPR) to hypothermia and rewarming (3), we theorized that incomplete activation of the endoplasmic reticulum associated degradation (ERAD) pathway could cause proteasome insufficiency and white matter injury. We randomized neonatal piglets to HI or sham surgery followed by overnight hypothermia (induced at 2 h), hypothermia + rewarming (0.5°C/h), or normothermia. Piglets recovered for 2, 6, 20, or 29 h. The number of subcortical white matter glial cells immunopositive for endoplasmic reticulum to nucleus signaling-1 (ERN1) at 2 h exceeded that of 29 h after HI and hypothermia ($p<0.05$; n=6-8). By western blotting, heat shock protein 70 (HSP70) levels at 6 h also exceeded that of 29 h after HI ($p<0.05$; n=4). However, the UPR markers p-PERK, p-ERN1, ATF6, and GADD34 remained similar to shams through 29 h after HI ($p>0.1$ for all comparisons; n=4). This response indicates an attenuated UPR after HI with incomplete ERAD. A consequence of impaired ERAD is failure to activate proteasomes that clear oxidized and damaged proteins after HI. We randomized separate piglets to HI or sham surgery followed by overnight hypothermia, hypothermia + rewarming (0.5°C/h), or normothermia. Immunoblotting revealed that proteasome 20S levels peaked at 20 h and then declined 29 h after HI, hypothermia, and rewarming ($p<0.05$; n=4). Carbonylated protein levels were higher at 29 h than at 20 h after HI and hypothermia ($p<0.05$; n=4). Ubiquitinated proteins levels were also higher 29 h after HI and hypothermia than those at 6 and 20 h after HI and hypothermia or normothermia ($p<0.05$ for each comparison; n=4). These data suggest that an attenuated UPR with incomplete ERAD leads to insufficient proteasome function after HI and hypothermia. Proteasome insufficiency is consistent with the accumulation of carbonylated and ubiquitinated proteins that are likely to be dysfunctional, potentially cytotoxic, and may cause white matter injury with oligodendrocyte apoptosis. Thus, proteasome activation has potential as an adjunct to hypothermia to mitigate white matter injury

in HIE.
1. Lee JK, et al. Dev Neurosci 2016; DOI: 10.1159/000452833.
2. Wang B, et al. Neuroscience 2016; 316:296-310.
3. Lee JK, et al. Dev Neurosci. 2016; 38:277-294.

Disclosures: P. Santos: None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.18/EE5

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: CIHR

RI MUHC Desjardins Studentship in Child Health Research

Heart and Stroke Foundation

Hoppenheim Fund

Fondation des Étoiles

The Montreal Children's Hopital Foundation

HSBC

Title: Added value of interleukin-1 blockade to hypothermia in neonatal encephalopathy due to inflammatory-sensitized hypoxia-ischemia: A preclinical study

Authors: *M. CHEVIN[1], G. SEBIRE[2]
[1]Pediatrics Dept., McGill Univ., Montreal, QC, Canada; [2]Pediatrics, MUHC, Montreal, QC, Canada

Abstract: Background: Neonatal encephalopathy (NE) and subsequent cerebral palsy (CP) resulting from hypoxia-ischemia (HI) or inflammatory-sensitized HI remain very prevalent and lead to significant mortality and morbidity. Few neuroprotective treatments are available against NE: they are limited to symptomatic care and hypothermia (HT), leaving about 50% of patients with neurological sequelae. HT is effective in alleviating NE in some, but not all, human term newborns. It has been reported that these newborns exposed to inflammatory-sensitized HI might have less therapeutic benefit from HT than those exposed to HI alone. Our work and others uncovered that interleukin-1 (IL-1) is at the apex of the inflammatory cascade generating brain injury, and that HT fails to counteract the IL-1 system in NE (Chevin *et al*., Int J Dev Neurosci,

2016). This supports a potential neuroprotective benefit of IL-1 receptor antagonist (IL-1Ra) as targeted add-on therapy to HT. Objective: We tested the added value of IL-1Ra administration to the neuroprotective effect of HT in NE. Methods: We used a rat model of lipopolysaccharide (LPS)+HI-induced NE at postnatal (P) day 12. Inflammation was induced by injecting intraperitoneally (ip) 50 µg/kg of LPS from *E.coli*. Four hours (h) later, the right common carotid artery was ligated, then hypoxia was induced (8% O_2, 1 h 30 min). Pups were submitted (or not) to HT (32° ± 0.5°C, 4 h). IL-1Ra (12.5 - 200 mg/kg q12 h) *vs* saline was injected ip from P12 to P14. Results: HT alleviated brain injury in the ischemic penumbra (neocortex and hippocampus), but not core injury, of LPS+HI-exposed pups. This neuroprotective effect did not result from a down-regulation of the neuroinflammatory response mediated by IL-1β or TNF-α. IL-1Ra treatment (50 mg/kg) was well tolerated. It reduced core injuries and mortality of LPS+HI+HT-exposed pups. Conclusion: Our results demonstrate that IL-1Ra (50mg/kg) has an added value to the neuroprotective effect of HT in LPS+HI-induced NE. This project could open new therapeutic avenues to prevent CP.

Disclosures: M. Chevin: None. **G. Sebire:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.19/EE6

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: Japan Society for the Promotion of Science Grant-in-Aid for Scientific Research(C) 16K11750

Title: Effects of dexmedetomidine on brain ischemia

Authors: *S. MAEDA[1], R. ONISHI[2], Y. HONDA-WAKASUGI[2], A. YABUKI-KAWASE[3], H. HIGUCHI[1], T. MIYAWAKI[2]
[1]Dept. of Dent. Anesthesiol. Okayama Univ.Hosp., Okayama-Shi, Japan; [2]Dept. of Dent. Anesthesiol. and Special Care Dent., [3]Ctr. for Promotion of Dent. Educ. and Intl. Collaboration, Okayama Univ. Grad. Sch. of Medicine, Dent. and Pharmaceut. Sci., Okayama, Japan

Abstract: Background: The number of anesthetic managements of elderly patients has increased along with progresses of anesthetics and monitoring of patients. One of the major problems in anesthetic management of elderly patients is post-operative cognitive impairment (POCI). Although direct causes for the POCI remain unclear, unstable hemodynamics and brain inflammation are likely to associated with the POCI. Dexmedetomidine, a selective agonist on alpha 2 adrenaline receptor, can pass through blood brain barrier and act on the receptor in the CNS, and it has been established to have an anti-inflammatory effect in periphery. Therefore, it

is thought to be a possible treatment to control the POCI. And in this study, effects of dexmedetomidine against an ischemia and an inflammation in the mouse brain were evaluated. Material and Methods: As the first experiment, the effects of dexmedetomidine was evaluated against mouse ischemia/reperfusion model, which is brought by selective occlusion of middle cerebral artery with modified monofilament through internal carotid artery. The effect was assessed by comparing infarct areas in brain section stained with triphenyltetrazolium chloride. In the second experiment, we made mouse model of brain ischemia with inflammation by a temporally occlusion of bilateral common carotid arteries and an injection of low dose lipopolysaccharide, and effects of dexmedetomidine was examined with immunostaining and realtime RT-PCR for items related with apotosis, inflammation and oxidative reaction. Results: In the first experiment, brain infarction was brought in the mouse model of brain ischemia/reperfusion. But, dexmedetomidine did not decrease the infarction area in the model although sedative effect of dexmedetomidine was shown clearly. Conclusion: Dexmedetomidine did not reduce the infarct area. However, it is likely effective against reaction in brain brought by hemodynamic changes and inflammation.

Disclosures: S. Maeda: None. **R. Onishi:** None. **Y. Honda-Wakasugi:** None. **A. Yabuki-Kawase:** None. **H. Higuchi:** None. **T. Miyawaki:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.20/EE7

Topic: H.01. Animal Cognition and Behavior

Support: FAPESP 2013/20378-8

Title: The effects of Ginkgo biloba standardized extract in the serotonergic, glutamatergic and gabaergic receptors modulation in the dorsal hippocampal formation (dHF) of rats submitted to acquisition of conditioned suppression

Authors: *M. TILGER, C. R. ZAMBERLAM, S. M. CERUTTI
Univ. Federal De São Paulo, Diadema, Brazil

Abstract: Recent studies from our lab have shown that EGb is able of modulating conditioned suppression by inducing differential expression of genes and proteins in the dHF of rats. Further, we have analyzed of gene expression in the dHF after conditioned suppression and treatment with flavonoidic fraction from Erythrina falcata and showed that NMDAR-GluN2B and 5-HT1ARs, $GABA_AR$ α1 and $GABA_AR$ α5 play an important role in synaptic plasticity and therefore contribute to the acquisition of fear memory. Furthermore, to better understand the effects of EGb and the contributions of these receptors as well as their interactions in the dHF

(CA3, CA1 and DG) were assessed for the first time by administering agonists, antagonists of receptors for GABA, glutamate (NMDA) and 5-HT or one these antagonists before EGb prior the conditioning session. Male adults Wistar rats (CEUA nº0268/12) were randomly assigned to the control (vehicle, Tween 80 12%) and EGb-treated groups (0.25, 0.5 ou 1 $g.Kg^{-1}$). After completing the behavioral tests the animals were deeply anesthetized and transcardially perfused with formalin 4% (n=3 per group). The encephalon was extracted and frozen at -80ºC. Coronal sections (20µm) were made with cryostat microtome. The slides contained the sections of dHF, were submitted to imunnohistochemical procedure for estimation of total GluN2B, 5-HT1ARs, $GABA_AR$ α1 and α5 and GFAP immuno-positive cells in the both side of the dHF and to imunnofluorescence of double labeling to colocalization of one these receptors with astrocytes (GFAP) or gabaergic interneurons (GAD-67). The images were acquired in AxioImagerA2 microscope with Zen 2 software, the imunnoreactive cells (IR) were quantified in four fields/region/hemisphere with ImageJ software, the statistical analyzes were made with one-way ANOVA and Tuckey post-hoc in the GraphPad Prism 6.0 software. We found that the treatment with EGb at doses 0.5 and 1.0 $g.Kg^{-1}$ increased the number of GluN2BR-IR cells and $GABA_AR$ α1-IR in the CA1, CA3 and DG of dHF. In addition, the treatment with 0.25 $g.Kg^{-1}$ EGb increases $5HT_{1A}R_{-IR}$ cells in the CA3 and DG ($p<0.05$) and treatment with 0.25 and 0.5 $g.Kg^{-1}$ EGb increased $GABA_AR$ α5-IR cells in the CA1 at the three doses after EGb in the CA3 and DG ($p<0.05$) in relation to control group. Furthermore, we found that all receptors, except $GABA_AR$ α1, were colocalizated with GAD-67 and GFAP except. Together, our data suggest that EGb modulates these receptors in a dose-dependent and region-specific manner and confirm our hypothesis that the hippocampus may be involved in suppressing the licking response and thereby be one of the targets of drug action in the modulation of conditioned suppression.

Disclosures: M. Tilger: None. **C.R. Zamberlam:** None. **S.M. Cerutti:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.21/EE8

Topic: B.12. Glial Mechanisms

Support: NRF 2016R1D1A1B01009186

NRF 2015R1A2A1A10051958

Title: EBP50 as a molecular link between ErbB2 signaling and peripheral neuropathy

Authors: *G. SONG[1], D. P. GUPTA[1], M. H. RAHMAN[1], H. PARK[2], K. CHUNG[3], B.-O. CHOI[4], K. SUK[1]
[1]Brain Sci. and Engin. Institute, Dept. of Pharmacology,, Kyungpook Natl. Univ., Daegu-City,

Korea, Republic of; [2]Dept. of Physiol., Col. of Med, Dong-A Univ., Pusan, Korea, Republic of; [3]Dept. of Biol. Sci., Kongju Natl. Univ., Gongju, Korea, Republic of; [4]Dept. of Neurol., Samsung Med. Center, Sungkyunkwan Univ. Sch. of Medicine., Seoul, Korea, Republic of

Abstract: The peripheral nervous system has remarkable regenerative capability. Regeneration in the peripheral nervous system depends upon the abilities of Schwann cells; de-differentiation, remyelination, and interaction with axon. Schwann cells-neuron interaction is known to be modulated via neuregulin/ErbB2 signaling but the underlying molecular mediators remain less well understood. Here, we introduced ERM binding protein 50 (EBP50) protein as a novel regulator of ErbB2-mediated Schwann cell functions. EBP50 is highly expressed at the tip of Schwann cells and the nodes of Ranvier, where the projections of the Schwann cells are perpendicular to. EBP50 expression in sciatic nerve significantly increased after crush injury, suggesting its functional role during injury-induced nerve degeneration/regeneration process. Knockdown of EBP50 in Schwann cells reduced the length of cell projections and cell motility. We further showed that EBP50 regulated the expression and localization of ErbB2 and the NRG-induced AKT activation in Schwann cells. In addition, the heterozygous knockout mice (EBP50+/-) have abnormal myelination and decreased motor function, showing some features of peripheral neuropathy. Finally, EBP50 heterozygote mutations were found in the patients with Charcot-Marie-Tooth neuropathy. Together, our results proposed that EBP50 is a key regulator of ErbB2-mediated peripheral nerve degeneration/regeneration.

Disclosures: G. Song: None. **D.P. Gupta:** None. **M.H. Rahman:** None. **H. Park:** None. **K. Chung:** None. **B. Choi:** None. **K. Suk:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.22/EE9

Topic: F.05. Neuroimmunology

Support: CNPq Grant 477421/2013-0

UFCSPA

Title: Obesity-induced neuroinflammation is decreased by omega-3 supplementation

Authors: *R. P. GUEDES[1], A. ANDRADE[1], B. PRAETZEL[1], M. PORAWSKI[1], L. S. DE FRAGA[2], M. GIOVENARDI[1]
[1]Fed. Univ. of Hlth. Sci. of Porto Alegre, Porto Alegre, Brazil; [2]Federal Univ. of Rio Grande Do Sul - UFRGS, Porto Alegre, Brazil

Abstract: Obesity is characterized by a systemic inflammatory status that affects multiple organs and body systems, including the central nervous system (SNC). Obesity induced-neuroinflammation generates deleterious effects in the SNC, including increased susceptibility to neurodegenerative diseases and cognitive impairment. Omega-fatty acids are well known for their anti-inflammatory and neuroprotective role. However, it is not known whether their supplementation can revert neuroinflammation. The present study aimed to evaluate the impact of omega-3 supplementation on neuroinflammatory markers of rats fed a high-fat diet. Male Wistar rats received standard diet (SD) or high fat diet (HFD) for 20 weeks. Omega-3 supplementation or vehicle administration started at 16th week. Omega-3 supplementation did not modify the total weight gain, but it lowered the visceral adiposity following HFD. We also found an improvement in insulin sensitivity following HFD plus omega-3. We also demonstrated that omega-3 supplementation decrease astrogliosis after HFD in the cerebral cortex through GFAP immunostaining. HFD increased gene expression of interleukin-6 and tumor necrosis factor-α in the cerebral cortex and omega-3 treatment decreased gene expression of these proinflammatory cytokines. Thus, our results demonstrate that omega-3 provides a beneficial metabolic role and a reduction of the neuroinflammatory profile following HFD, reinforcing the anti-inflammatory role of this family of fatty acids.

Disclosures: R.P. Guedes: None. **A. Andrade:** None. **B. Praetzel:** None. **M. Porawski:** None. **L.S. De Fraga:** None. **M. Giovenardi:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.23/EE10

Topic: F.05. Neuroimmunology

Title: Calcium release-activated calcium (CRAC) channel inhibiton protects against experimental stroke

Authors: *A. MIZUMA[1], R. KACIMI[2], K. STAUDERMAN[3], M. J. DUNN[3], S. HEBBAR[3], M. A. YENARI[2]
[1]Neurol., Univ. of California, San Francisco and Vetera, San Francisco, CA; [2]Univ. of California, San Francisco & San Francisco Veterans Affairs Med. Ctr., San Francisco, CA; [3]CalciMedica, La Jolla, CA

Abstract: Purpose: Inflammatory responses after ischemic stroke contribute to worsening of ischemic brain injury. Calcium release-activated calcium (CRAC) channels are known to be involved in the activation of immune responses in microglia, and have been studied indirectly through non-specific calcineurin inhibitors such as cyclosporine and FK506. Since these compounds have many off-target effects and unacceptable side effects, we studied a novel

CRAC channel inhibitor which is specific to the CRAC channel and appears safe in preclinical studies in anticipation of human clinical trials for other indications. We explored whether this inhibitor might improve outcome in experimental stroke by inhibiting microglial activation.
Subjects and Methods: 2-3 month old C57/BL6 male mice, were exposed to transient middle cerebral artery occlusion (tMCAO, filament model) for 2h followed by reperfusion. Some were treated with the CRAC channel inhibitor (CM-EX-137, 5mg/kg IP; tMCAO-CM) or vehicle control (tMCAO) daily beginning immediately after stroke onset. Brains were collected 3d post-stroke. Neurological function was evaluated using a Bederson score, elevated body swing test, adhesive removal test, and corner test at baseline, at 3, 24 and 72 h. Relative cerebral blood flow (rCBF) was studied in some mice using laser Doppler flowmetry. Ischemic lesion size was evaluated by 2,3,5-triphenyl tetrazolium chloride (TTC) staining. Microglial/monocyte activation was assessed by isolectin B4 histochemistry.
Results: A total of 18 mice were studied (tMCAO-CM: 8; tMCAO: 8, sham:2). 3 mice exposed to tMCAO (1 tMCAO-CM; 2 tMCAO) died before study completion (differences in mortality N.S). rCBF was no different between experimental groups, and is consistent with the notion that the compound should not alter blood flow. Neurological function at 1 & 3d after ischemia was significantly improved in
tMCAO-CM group compared to tMCAO ($p<0.001$). The CM compound also significantly reduced infarct volume by ~60% ($p<0.05$). Isolectin B4 staining was markedly reduced in the tMCAO-CM group compared to tMCAO 3d post ischemia ($p<0.01$).
Conclusion: CRAC channel inhibition protects against experimental stroke by decreasing lesion size, improving neurological outcomes and decreasing microglial activation.

Disclosures: A. Mizuma: None. **R. Kacimi:** None. **K. Stauderman:** None. **M.J. Dunn:** None. **S. Hebbar:** None. **M.A. Yenari:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.24/EE11

Topic: B.12. Glial Mechanisms

Support: MH1R01MH093362

T32 NS 007453

Title: Delineating mechanisms underlying panic pathophysiology

Authors: *A. WINTER[1], R. AHLBRAND[1], L. VOLLMER[1], N. QUAN[2], E. G. KRAUSE[3], R. SAH[1]

[1]Psychiatry, Univ. of Cincinnati, Cincinnati, OH; [2]Dept Biosci., Ohio State Univ., Columbus, OH; [3]Dept Pharmacodynamics, Univ. of Florida, Gainesville, FL

Abstract: Panic disorder (PD) is a prevalent, complex anxiety disorder afflicting about 3% of U.S. adults. The hallmark of PD is recurring panic attacks; episodes of debilitating fear concurrent with intense physical symptoms. Existing treatments for PD are limited, and many patients are unresponsive. This highlights the need for improved mechanistic understanding of the biological triggers of panic attacks to identify novel targets of intervention. Panic attacks can occur spontaneously without external threats, suggesting internal homeostatic disturbances as likely triggers. Accordingly, PD patients are very sensitive to challenges producing pH imbalances, specifically acidosis. Carbon dioxide (CO_2) inhalation induces acidosis and reliably elicits panic attacks in PD patients. Thus, analysis of acid-sensing mechanisms and downstream effectors are crucial to understanding panic pathophysiology. Consistent with the negative valence construct within NIMH's Research Domain Criteria initiative, our lab employs CO_2 inhalation in mice to simulate an acute, interoceptive threat producing defensive behaviors representative of fear. Recently, we reported contributions of microglial acid-sensing to CO_2-evoked fear (Vollmer, 2016), however specific sites and neuronal effector mechanisms remain unclear. The current study a) confirms the brain site orchestrating acidosis-evoked behavior and b) identifies a potential neuronal coupling system in CO_2 responses: the renin angiotensin system (RAS). Site directed infusion of acidified aCSF into a homeostatic regulatory region, the subfornical organ (SFO), evoked significant freezing in mice dependent on the presence of the acid sensor. This suggests the SFO plays a pivotal role in regulating PD-relevant behavioral responses to acidosis. The SFO is also a known modulator of systemic homeostasis via RAS. To investigate the role of RAS in our CO_2 inhalation model, Angiotensin II-type I receptor antagonist, losartan, was delivered intracerebroventricularly near the SFO. Significant attenuation of CO_2-evoked freezing was observed in losartan treated mice, identifying RAS as a potential neuronal effector downstream of microglial acid sensing. Ongoing studies are characterizing microglial-neuronal coupling at functional and cellular levels using transgenic mice and DREADDs. Preliminary evidence suggests microglia-blood vessel-neuronal interactions, outlining a novel tripartite mechanism in CO_2-evoked adaptive fear responses relevant to PD. Our studies help delineate mechanisms by which homeostatic triggers may lead to panic attacks, and will identify targets for future therapeutic intervention.

Disclosures: **A. Winter:** None. **R. Ahlbrand:** None. **L. Vollmer:** None. **N. Quan:** None. **E.G. Krause:** None. **R. Sah:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.25/EE12

Topic: B.12. Glial Mechanisms

Support: SFB 1158

Title: GABAergic depolarization in C-fiber axons is modulated directly and indirectly by allopregnanolone via PKC_e

Authors: V. BONALUME[1], L. CAFFINO[1], R. W. CARR[2], L. F. CASTELNOVO[1], D. COLLEONI[1], S. MELFI[1], F. FUMAGALLI[1], *M. SCHMELZ[2], V. MAGNAGHI[1]
[1]Dept. of Pharmacol. and Biomolecular Sci., Milan Univ., Milano, Italy; [2]Heidelberg Univ., Mannheim, Germany

Abstract: Schwann cells (SC) are specialized glial cells of the peripheral nervous system (PNS) some of which forming the myelin sheath and interacting functionally with neurons under basal and pathophysiological conditions. Schwann cells express ionotropic GABA-A and metabotropic GABA-B receptors and are able to synthetize both GABA and the neuroactive steroid allopregnanolone (ALLO), a progesterone metabolite known to bind selectively to GABA-A receptors.
It is well established that ALLO and GABA-A activation can regulate SC in the PNS in terms of differentiation, myelination and regeneration. However, the function of GABA-A receptors in peripheral axons and their role in SC-neuron cross-talk is still unclear.
To this end, we performed an electrophysiological characterization of GABA-A receptors on unmyelinated C-fibers in mouse sural nerve. GABA is commonly associated with inhibitory, hyperpolarizing effects in the central nervous system. However, GABA-A receptor activation resulted in axonal depolarization and increased excitability in peripheral C-fibres. This effect was blocked by bicuculline, mimicked by muscimol and enhanced by benzodiazepines and ALLO (1 μM) which produced a leftward shift of the dose-response curve. In addition to this rapid modulation of C-fibres, ALLO can also control intracellular pathways in neurons via SCs. Indeed, we observed that neurons exposed to conditioned medium of SCs treated with ALLO (1 μM) exhibit an upregulation of the ε isoform of protein kinase C (PKCε), an intracellular pathway known to be involved in the pathogenesis and maintenance of neuropathic pain conditions. The mediator responsible for this neuronal-glia modulation might be Brain-Derived Neurotrophic Factor via TrkB. Treatment of SCs with ALLO, indeed, results in the production and release of BDNF. In neurons, conditioned medium mediated up-regulation of PKCε was mimicked by recombinant BDNF and blocked by a TrkB inhibitor.
In conclusion, we suggest that SC and peripheral neurons interact in two ways: SC-derived ALLO can acutely sensitize GABA-A receptors modulating axonal excitability and SC-derived BDNF can lead to long-term upregulation of neuronal PKCε.

Disclosures: V. Bonalume: None. **L. Caffino:** None. **R.W. Carr:** None. **L.F. Castelnovo:** None. **D. Colleoni:** None. **S. Melfi:** None. **F. Fumagalli:** None. **M. Schmelz:** None. **V. Magnaghi:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.26/EE13

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Title: Roles of autophagy in palmitate-induced ER stress and apoptosis in hypothalamic neuronal cells

Authors: *Y. LIM, E.-K. KIM
DGIST, Daegu, Korea, Republic of

Abstract: Autophagy is a cellular process that degrades damaged organelles and aggregated proteins. Hypothalamic autophagy is important for regulation of food intake and energy expenditure. High-fat diet induces hypothalamic injury through endoplasmic reticulum (ER) stress and apoptosis in rodents. We demonstrate here that autophagy plays a role in protecting hypothalamic neuronal cells from ER stress-induced apoptosis. Treatment of palmitate induces ER stress and autophagy in hypothalamic neuronal cells, N41. However, prolonged treatment of palmitate impairs autophagy and increases apoptosis. When palmitate-induced autophagy is enhanced by rapamycin, an autophagy inducer, ER stress and apoptosis are decreased. On the other hand, treatment of bafilomycin, an autophagy inhibitor, increases ER stress and apoptosis. In addition, pretreatment of ER stress reducer to palmitate decreases both autophagy and apoptosis. Taken together, our results indicate that autophagy has a protective role in cellular homeostasis by regulating ER stress. Our study suggests that modulation of autophagy is a possible therapeutic strategy for metabolic disorders such as obesity and diabetes.

Disclosures: **Y. Lim:** None. **E. Kim:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.27/EE14

Topic: C.06. Neurotoxicity, Inflammation, and Neuroprotection

Support: NRF Grant NRF-2015R1A6A3A01060724

NRF Grant NRF-2014R1A2A1A11054095

NRF Grant NRF-2015R1A4A1042271

RDA Grant PJ01179202

KRIBB Grant KGM4611613

Title: Peroxiredoxin 5 decreases beta-amyloid-mediated cyclin-dependent kinase 5 activation through regulation of Ca^{2+}-mediated calpain activation

Authors: *J. SEONG[1], *H. J. LEE[2], D.-S. LEE[1]
[1]BK21 Plus KNU Creative Bioresearch Group, Kyungpook National University, Daegu, Korea, Republic of; [2]Chung-Ang Univ. Hosp., Biomed. Res. Inst., Seoul, Korea, Republic of

Abstract: AIMS:Aberrant Cdk5 (cyclin-dependent kinase 5) and oxidative stress are crucial components of diverse neurodegenerative disorders, including Alzheimer's disease (AD). We previously reported that a change in peroxiredoxin (Prx) expression is associated with protection from neuronal death. The aim of the current study was to analyze the role of Prx in regulating Cdk5 activation in AD.RESULTS:We found that of the six Prx subtypes, Prx5 was increased the most in cellular (N2a-APPswe cells) model of AD. Prx5 in the brain of APP (amyloid precursor protein) transgenic mouse (Tg2576) was more increased than a nontransgenic mouse. We evaluated Prx5 function by using overexpression (Prx5-WT), a mutation in the catalytic residue (Prx5-C48S), and knockdown. Increased neuronal death and Cdk5 activation by amyloid beta oligomer (AβO) were rescued by Prx5-WT expression, but not by Prx5-C48S or Prx5 knockdown. Prx5 plays a role in Cdk5 regulation by inhibiting the conversion of p35 to p25, which is increased by AβO accumulation. Prx5 is also upregulated in both the cytosol and mitochondria and it protects cells from AβO-mediated oxidative stress by eliminating intracellular and mitochondrial reactive oxygen species. Moreover, Prx5 regulates Ca2+ and Ca2+-mediated calpain activation, which are key regulators of p35 cleavage to p25. Innovation and Conclusion: Our study represents the first demonstration that Prx5 induction is a key factor in the suppression of Cdk5-related neuronal death in AD and we show that it functions via regulation of Ca2+-mediated calpain activation.

Disclosures: H.J. Lee: None. **D. Lee:** None.

Poster

396. CNS and PNS Injury and Therapeutics

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 396.28/EE15

Topic: G.05. Anxiety Disorders

Support: Fapemig

Capes

CNPQ

UFOP

Title: Influence of pulmonary inflammation on behavior in wistar rats exposed to cigarette smoke

Authors: *M. T. CHÍRICO, S. I. S. R. NORONHA, G. S. V. CAMPOS, P. M. A. LIMA, M. R. GUEDES, A. B. F. SOUZA, F. C. S. SILVA, A. B. FIGUEIREDO, L. C. C. AFONSO, S. D. CANGUSSÚ, D. A. CHIANCA-JR, F. S. BEZERRA, R. C. A. MENEZES
Univ. Federal De Ouro Preto, Ouro Preto, Brazil

Abstract: Background - Cigarette smoke (CS) is a complex combination that contains more than 7,000 chemicals substances, which can promote oxidative damage to the lung parenchyma and induce inflammatory process. Pulmonary inflammation leads to neurons activation in the central nervous system, however, the neural mechanisms implicit and the relationship between lung inflammation and behavioral assessment are not clear. Our aim in this study was to evaluate inflammation the anxiety behavior of animals exposed to cigarette smoke with and without tobacco. Methods - Twenty-four male Wistar rats were divided into three groups: control group (CG); common cigarette (CC) and paper cigarette (CP), without tobacco. The animals were exposed to twelve cigarettes per day in three periods (morning, afternoon and night) for eight consecutive days. In addition to CS exposure, the animals were submitted to behavioral tests: Elevated T-maze (ETM) and open field (OF). The animals were euthanized for removal of bronchoalveolar lavage (BAL), blood and lung. LBA was used for total count, cell differential and for the measurement of pro-inflammatory cytokines; Blood plasma was analyzed for cytokine dosing; And the lung was used for dosage of the total Proteins, Glutathione system, antioxidant enzymes: SOD and CAT, TBARS and carbonylated protein. Results - Exposure to CC smoke reduced anxiety and panick-like behaviors. On the other hand, CP induced panic and anxiety behaviors. Animals exposed to common and paper CS showed an increase on the influx of inflammatory cells (CC: 170 ± 3.162 x 103/ml and CP: 180 ± 6.547 x 103/ml vs. CG: 118 ± 6.665 x 103/ml); (CC: 34.86 ± 1.855 U/mg prot and CP: 40.65 ± 3.444 U/mg prot vs. CG: 25.73 ± 1.092 U/mg prot) and CAT (CC : 0.96 ± 0.06 U/mg prot and CP: 0.94 ± 0.10 U/mg prot vs. CG: 0.68 ± 0.10 U/mg prot). The exposure also generated lung damage as demonstrated by increased TBARS levels (CC: 1.39 ± 0.15 nM/mg prot and CP: 1.54 ± 0.10 nM/mg prot vs. GC: 1.02 ± 0.05 nM/mg prot) and reduction of the ratio GSH/GSSG (CC: 4.016 ± 0.5503 μM and CP: 3.316 ± 0.8901 μM vs. GC: 7.283 ± 0.6019 μM). In addition to the stereological analysis of the lung sections, which also showed alteration of histoarchitecture by increasing the volume of the alveolar space (CC: 57.46 ± 1.92%/mm² and CP: 53.40 ± 1.83%/mm² Vs. GC: 43.36 ± 3.46%/mm²) and alveolar septal volume reduction (CC: 41.60 ± 1.95%/mm² and CP: 46.21 ± 1.85%/mm² vs. GC: 55.28 ± 3.52%/mm²) in the exposed animals. Conclusions - Exposure to common cigarette smoke as well as paper cigarettes (without tobacco) led to an inflammatory process and lung damage. Importantly exposure to smoke from CP led to a increase in panic and anxiety behaviors.

Disclosures: **M.T. Chírico:** None. **S.I.S.R. Noronha:** None. **G.S.V. Campos:** None. **P.M.A. Lima:** None. **M.R. Guedes:** None. **A.B.F. Souza:** None. **F.C.S. Silva:** None. **A.B. Figueiredo:** None. **L.C.C. Afonso:** None. **S.D. Cangussú:** None. **D.A. Chianca-Jr:** None. **F.S. Bezerra:** None. **R.C.A. Menezes:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.01/EE16

Topic: D.03. Somatosensation: Pain

Title: HTR2B in sensory neurons mediates pruritus to SSRI antidepressants

Authors: *S. LEE[1], P. CHO[2], J. JANG[3], D. P. ROBERSON[4], R. TONELLO[1], S. HWANG[5], S. HWANG[6], R. H. LAMOTTE[7], C. PARK[8], S. JUNG[9], T. BERTA[1]
[1]Anesthesiol., Univ. of Cincinnati, Cincinnati, OH; [2]Physiol. Lab., Seoul, Korea, Republic of; [3]2Department of Biomed. Science, Grad. Sch. of Biomed. Sci. Hanyang Univ., Seoul, Korea, Republic of; [4]Children's Hospital, Boston, Cambridge, MA; [5]4Department of Biomed. Sciences, Korea Univ. Col. of Med., Seoul, Korea, Republic of; [6]Hanyang Univ., Seoul, Korea, Republic of; [7]Dept Anesthesiol, Yale Univ. Sch. Med., New Haven, CT; [8]Dept. of Physiol., Gachon University, Col. of Med., Incheon, Korea, Republic of; [9]Hanyang Univ. Med. Sch., Seoul, Korea, Republic of

Abstract: Acute and chronic pruritus is frequently associated with skin, liver and kidney diseases, as well as viral infection and drug treatment. Antidepressants called selective serotonin reuptake inhibitors (SSRIs) are among the most common drugs prescribed in U.S. today and are well-known to elicit skin adverse reactions such as rashes, urticaria and pruritus, often leading to non-compliance and interruption of otherwise successful therapies. We found that functional serotonin receptor 2B (HTR2B), a G protein-coupled receptors, was expressed in pruriceptive primary sensory neurons and specifically mediated pruritus to various SSRI antidepressants, but not other common pruritogen including histamine or the anti-malaria drug chloroquine. Notably, mice injected with sertraline (Zoloft®) or fluoxetine (Prozac®) or citalopram (Celexa®) scratched immediately and intensively for 10 minutes, a scratching that is significantly attenuated by the specific HTR2B antagonist RS127445. Although HTR2B is expressed and functional in a particular subpopulation of TRPV1-expressing neurons that innervate the skin and are linked to itch, HTR2B induced neuronal activity and itch through the transient receptor potential channel 4 (TRPC4) and not through the most common and well-studied TRPV1, TRPV4 or TRPA1. Finally, sertraline-evoked itch was significantly attenuated by the specific TRPC4 blocker ML204, as well as in *Trpc4-/-* mice. Our data clearly demonstrates that HTR2B and TRPC4 expressed in pruriceptive neurons mediate itch to SSRI antidepressants revealing previously

unknown itch signaling and suggesting new avenues for preventing cutaneous side effects to SSRI antidepressants.

Disclosures: **S. Lee:** None. **P. Cho:** None. **J. Jang:** None. **D.P. Roberson:** None. **R. Tonello:** None. **S. Hwang:** None. **S. Hwang:** None. **R.H. LaMotte:** None. **C. Park:** None. **S. Jung:** None. **T. Berta:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.02/EE17

Topic: D.03. Somatosensation: Pain

Support: National Science Foundation of China #81271239 (C.M.)

IBMS/CAMS Dean's Fund #2011RC01 (C.M.)

the PUMC Youth Fund (X.S., and C.M.)

the PUMC Innovation Fund for Postgraduate #2016 (F.L.)

the CAMS Innovation Fund for Medical Sciences (CIFMS)

Title: Fc-epsilon receptor I in primary nociceptors as a Fc-epsilon receptor I in primary nociceptors as a sensor of IgE-immune complex may contribute to allergic ocular pruritus

Authors: ***F. LIU**[1,2], T. WANG[1], B. YUAN[1], C. MA[1]
[1]Dept. of Anatomy, Histology and Embryology, Inst. of Basic Med. Sciences, Chinese Acade, Beijing, China; [2]Dept. of Anatomy, Histology and Embryology, Sch. of Basic Medicine, Peking Union Med. Col., Beijing, China

Abstract: Ocular pruritus is the major symptoms of allergic conjunctivitis, which usually accompanied with an elevated serum level of antigen-specific IgE. Fc-epsilon receptor I (FcεRI),typically expressed on the immune cells and bind to the Fc region of antigen-specific immunoglobulin E (IgE), is the only high-affinity activating cell-surface receptor that plays a key role in allergic responses. Fc receptors such as high-affinity IgG receptor (FcγRI) and FcεRI were recently discovered on neurons of dorsal root ganglion (DRG) and trigeminal ganglion (TG). Previous studies demonstrated that IgG-immune complex (IgG-IC) can directly activate primary nociceptor of DRG. In this study, we investigated the potential role of neuronal FcεRI in ocular pruritus. We first established that the mouse demonstrated a remarkable wiping toward the eye with the forelimb when the algogen capsaicin was dropped to the eye, but showed a remarkable scratching toward the eye with hindlimb when a pruritogen Bam8-22 was applied to

the eye. Next, we induced a mouse model of ocular pruritus via topical application of antigen to the eyes in the mice sensitized with the allergen (oval albumin, OVA) together with the adjuvant (aluminum hydroxide). Challenging with OVA (0.1%-5%, 5μL dropped to the eye) induced dose-dependent scratching behavior, which cannot be fully blocked by pre-treatment of mast cell stabilizer (cromolyn sodium) or histamine I receptor antagonist (terfenadine). However, pre-treatment of FcεRI blocking antibody almost completely abolished the OVA-evoked ocular scratching. Futhermore, we found that all three subunits (FcεRIα, β and γ) are expressed on TG neurons that co-expressed with itch-specific neuronal makers such as MrgprA3 or MrgprD. The expression levels of FcεRIα and β subunits were found upregulated in the TG of sensitized mice. In acutely dissociated TG neurons, IgE-IC produced an increase in the intracellular calcium level and induced hyperexcitability in small-sized nociceptive TG neurons. The IgE-IC induced calcium increase could be knocked down with a specific FcεRIα siRNA in dissociated TG neurons. These results indicate that primary nociceptive neurons express functional FcεRI and could be directly activated by IgE-IC. Neuronal FcεRI may serve as the sensor to IgE immune complex and play an important role in allergic ocular pruritus.

Disclosures: **F. Liu:** None. **T. Wang:** None. **B. Yuan:** None. **C. Ma:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.03/EE18

Topic: D.03. Somatosensation: Pain

Title: Specialized mechanosensory nociceptors mediating rapid responses to hair-pull

Authors: *N. GHITANI[1], A. BARIK[1], M. SZCZOT[1], J. H. THOMPSON[1], C. LI[2], C. E. LE PICHON[3], M. J. KRASHES[2], A. T. CHESLER[1]
[1]NCCIH, [2]NIDDK, [3]NICHD, Natl. Inst. of Hlth. (NIH), Bethesda, MD

Abstract: The somatosensory system provides animals with the ability to detect, distinguish and respond to diverse thermal, mechanical and irritating stimuli. While there has been progress in defining classes of neurons underlying temperature sensation and gentle touch, less is known about the neurons specific for mechanical pain. Here, we use in vivo functional imaging to identify a class of cutaneous sensory neurons that are selectively activated by high threshold mechanical stimulation (HTMRs). We show that their optogenetic excitation evokes rapid protective and avoidance behaviors. Unlike other nociceptors, these HTMRs are fast conducting Aδ-fibers. Notably, we find that Aδ-HTMRs innervate unique but overlapping fields and can be activated by stimuli as precise as the pulling of a single hair. Together the distinctive features of this class of Aδ-HTMRs appear optimized for accurate and rapid localization of mechanical pain.

Disclosures: **N. Ghitani:** None. **A. Barik:** None. **M. Szczot:** None. **J.H. Thompson:** None. **C. Li:** None. **C.E. Le Pichon:** None. **M.J. Krashes:** None. **A.T. Chesler:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.04/EE19

Topic: D.03. Somatosensation: Pain

Support: NIH Grant 1 R01 AR070875-01

Blaustein Pain and Research Education Fund, Johns Hopkins University

Neurosurgery Pain Research Institute, Johns Hopkins University

Title: The MrgprD agonist β-alanine and bovine adrenal medullary protein 8-22, an MrgprX1 agonist, preferentially activate subtypes of polymodal nociceptive C-fibers in pigtail monkeys

Authors: A. H. KLEIN[1,2], M. WOOTEN[2], T. V. HARTKE[2], G. WU[2], *M. RINGKAMP[3]
[1]Pharm. Practice and Pharmaceut. Sci., Univ. of Minnesota Duluth, Duluth, MN; [2]Dept of Neurosurg., Johns Hopkins Univ., Baltimore, MD; [3]Dept Neurosurg., Johns Hopkins Univ. Dept. of Neurol. and Neurosurg., Baltimore, MD

Abstract: In mouse DRG, the expression of mas-related G protein-coupled receptors (Mrgprs) differentiates classes of polymodal nociceptive C-fiber neurons, that have been proposed to mediate distinct behaviors. MrgprD neurons have been shown to be involved in paw withdrawal to mechanical stimuli and scratching behavior induced by β-alanine, and MrgprA3 neurons have been proposed to represent a labeled line for itch. In primate, orthologous genes for some Mrgs exist (MrgD-G), whereas others, designated as MrgX1-4, cannot be clearly assigned to any of the MrgA-C subfamilies described in mice. Currently, it is unclear whether functionally distinct subclasses of cutaneous nociceptive C- fiber neurons expressing different Mrgprs can be differentiated in primate. Two types of heat responses are observed in cutaneous polymodal nociceptive C-fibers when their receptive fields (RF) are exposed to a stepped heat stimulus (49°C, 3s): a quick response (QCs) or a slow response (SCs). We have previously shown that QCs are preferentially activated by the MrgprD agonist β-alanine (ALA). Whether QCs and SCs also differ in their responses to bovine adrenal medullary protein 8-22 (BAM8-22), an Mrgpr agonist that activates murine MrgprC11 and human MrgprX1, is not known. Using standard teased-fiber techniques, neuronal activity of unmyelinated C fibers innervating the hairy skin was recorded in anaesthetized nonhuman male primates (Macaca nemestrina). After locating the mechanical RF of the fiber under study, sensitivity to noxious heat was tested using a contact free, temperature-controlled CO_2 laser system. Two blocks of intradermal injections (each 10 µl)

were then administered at the RF: one block consisted of extracellular fluid (ECF, the solvent) followed by ALA (90 µg) and another block of BAM8-18 (the inactive truncated peptide, 1 µg) followed by BAM8-22 (1 µg). The order of the blocks was randomized. Following each injection, neuronal activity was recorded for at least 5 minutes. A total of 45 C fibers was tested. Of 24 QCs, 23 responded to ALA and 17 responded to both, ALA and BAM8-22. QC- responses to ALA were about 2-fold larger than responses to BAM8-22. All 21 SCs responded to BAM8-22, but only 4 also responded to ALA. SC responses to BAM8-22 were about 20-fold higher than those induced by ALA. ALA responses in QCs were about 10-fold larger than in SCs, whereas BAM8-22 responses were about 3-fold larger in SCs than QCs. These results show that QCs and SCs are preferentially activated by ALA and BAM8-22, respectively, and suggest that QCs may represent neurons that preferentially express MrgprD whereas SCs may represent MrgprX1 expressing neurons.

Disclosures: A.H. Klein: None. **M. Wooten:** None. **T.V. Hartke:** None. **G. Wu:** None. **M. Ringkamp:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.05/EE20

Topic: D.03. Somatosensation: Pain

Support: Dean's Seed Fund of the RWTH Aachen University

German Israeli Foundation

Johannes and Frieda Marohn-Foundation

DFG grant GRK2162

Humboldt Foundation

Ministry for Innovation, Science and Research of North Rhine-Westphalia

Title: Rethinking neuronal excitability in pain: Nav1.7 contributes to action potential threshold, but not subthreshold depolarizations, in patient iPS cell-derived nociceptors

Authors: *J. E. MEENTS[1], E. BRESSAN[1], S. SONTAG[2,4], A. FOERSTER[1], P. HAUTVAST[1], M. HAMPL[1,5], H. SCHÜLER[3], R. GOETZKE[4], T. K. C. LE[1], I. P. KLEGGETVEIT[6], K. LE CANN[1], Z. KOHL[7], M. SCHMELZ[8], W. WAGNER[4], E. JORUM[6,9], B. NAMER[5], B. WINNER[10], M. ZENKE[2,4], A. LAMPERT[1]
[1]Inst. of Physiol., [2]Inst. for Biomed. Engin., [3]Inst. of Human Genet., Uniklinik RWTH Aachen,

Aachen, Germany; [4]Helmholtz-Institute for Biomed. Engin., RWTH Aachen Univ., Aachen, Germany; [5]Inst. for Physiol. and Pathophysiology, Friedrich-Alexander-University Erlangen-Nürnberg, Erlangen, Germany; [6]Dept. of Neurol., Oslo Univ. Hospital-Rikshospitalet, Oslo, Norway; [7]Univ. Hosp. Erlangen, Erlangen, Germany; [8]Heidelberg Univ., Mannheim, Germany; [9]Inst. of Clin. Med., Univ. of Oslo, Olso, Norway; [10]FAU Erlangen-Nuernberg, Erlangen, Germany

Abstract: The chronic pain syndrome erythromelalgia (IEM) is attributed to mutations in the voltage-gated sodium channel (Nav) 1.7. Here, we differentiated induced pluripotent stem cells (iPSC) from IEM patients with the Nav1.7/I848T mutation into nociceptors. Action potentials in these IEM nociceptors displayed a decreased firing threshold, an enhanced upstroke and a stronger hyperpolarization, all of which can explain the increased pain experienced by patients. Furthermore, IEM nociceptors showed a hyperpolarized Nav activation compared to unmutated controls. Application of the Nav1.7 inhibitor ProTx-II resulted in a loss of the mutation-induced shift of activation and revealed a hyperpolarized Nav activation in unmutated neurons. We conclude that Nav1.7 is not active during subthreshold depolarizations. Instead, its activity defines the action potential threshold and contributes significantly to the action potential upstroke. Thus, our findings may change our current understanding of the role of human Navs in the generation of action potentials and open new avenues for pharmacological intervention in pain syndromes.

Disclosures: J.E. Meents: None. **E. Bressan:** None. **S. Sontag:** None. **A. Foerster:** None. **P. Hautvast:** None. **M. Hampl:** None. **H. Schüler:** None. **R. Goetzke:** None. **T.K.C. Le:** None. **I.P. Kleggetveit:** None. **K. Le Cann:** None. **Z. Kohl:** None. **M. Schmelz:** None. **W. Wagner:** None. **E. Jorum:** None. **B. Namer:** None. **B. Winner:** None. **M. Zenke:** None. **A. Lampert:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.06/EE21

Topic: D.03. Somatosensation: Pain

Title: The role of Nav1.9 in somatosensory perception

Authors: *J. SALVATIERRA[1], X. DONG[2], F. BOSMANS[1]
[1]Physiol., [2]Neurosci., Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: The somatosensory nervous system perceives and transfers sensory modalities such as pain, itch, touch, and temperature from the periphery to the central nervous system. Voltage-gated sodium (Na_v) channels are vital for action potential generation, and are key players in this

signal transmission process. Out of the nine Na_v channel subtypes, $Na_v1.7$, $Na_v1.8$, and $Na_v1.9$ are particularly important for the transmission of sensory modalities due to their strategic expression in the somatosensory nervous system. Although $Na_v1.7$ and $Na_v1.8$ have been extensively studied with respect to specific modalities, the role of $Na_v1.9$ remains unclear. However, accumulating evidence suggests a prominent role for this channel subtype in nociception. Challenges in expressing this channel in a heterologous system and a poor understanding of its tissue distribution complicate the formation of a hypothesis about the role of $Na_v1.9$ in transmitting sensory modalities. To overcome these obstacles, we developed a $Na_v1.9$ stable cell line and generated an EGFP-tagged $Na_v1.9$ mouse line, which allows us to refine tissue distribution and function of this channel in particular cells. For example, we took advantage of our EGFP-tagged $Na_v1.9$ mice by performing fluorescent activated cell sorting (FACS) of dorsal root ganglion (DRG) neurons. Fluorescent cells were subjected to transcriptional profiling which revealed a multitude of genes that were differentially expressed in $Na_v1.9$-expressing neurons, including several G-protein coupled receptors (GPCRs) implicated in somatosensory perception. We are currently using known ligands and mouse lines for these GPCRs to investigate their link with $Na_v1.9$. Our goal is to determine how modulation of $Na_v1.9$ via these GPCRs alters somatosensory perception.

Disclosures: **J. Salvatierra:** None. **X. Dong:** None. **F. bosmans:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.07/EE22

Topic: D.03. Somatosensation: Pain

Support: MOST 105-2320-B-008-003

Title: Proton-sensing GPCRs, G2A and OGR1, participate in establishing hyperalgesic priming

Authors: C.-W. LEE, *W.-H. SUN
Natl. Central Univ., Jhongli, Taoyuan, Taiwan

Abstract: High local concentration of protons was found in kinds of lesions and injury, such as fibromyalgia, immune disease or arthritis, accounted for part of chronic pain. Tissue acidosis triggers a signal switch (2-4hr after acid injection) from Gs-protein kinase A (PKA) to Gi-protein kinase Cε (PKCε) and the PKA-to-PKCε signal switch is associated with development of hyperalgesic priming. Proton-sensing G-protein-coupled receptors including OGR1, G2A, TDAG8, and GPR4, could be major candidates to mediate the PKA-to-PKCε signal switch. We have previously found hat T-cell death-associated gene 8 (TDAG8) mediates acidosis signals and couples to Gs protein to activate PKA pathway, initiating hyperalgesia. TDAG8-Gs-PKA

pathway also participates in the establishment of hyperalgesic priming. However, the mechanism underlying the PKA-to-PKCε signal switch remains unclear. In this study, we used shRNA knockdown technique to explore the transition from acute to chronic pain. In the dual acid injection model (acid [pH5.0] injected twice, 5 days apart), co-injection of G2A- and OGR1-shRNA plasmids significantly inhibited the prolonged phase (after 4hr) of the first hyperalgesia and shortened the duration of the second hyperalgesia, which corresponds to the action time of the Gi-PKCε signaling. Given that a heteromer of G2A and OGR1 mediates Gi-signaling, G2A and OGR1 may participate in the establishment of hyperalgesic priming through regulation of the Gi-PKCε signaling.

Disclosures: C. Lee: None. **W. Sun:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.08/EE23

Topic: D.03. Somatosensation: Pain

Support: Instituto Carlos III, FIS PI14/00141

Instituto Carlos III, RETIC RD12/0034/0003 and RD16/0008/0014

Ministerio de Economia y Competitividad, BFU2014-56572-P

Generalitat de Catalunya, 2014SGR1165

Ministerio de Economia y Competitividad, RYC2011-08589

Title: Pyrethroids inhibit K_{2P} channels and activate sensory neurons: Basis of insecticide-induced paresthesias

Authors: A. CASTELLANOS[1,2], A. ANDRES[1], L. BERNAL[3], G. CALLEJO[1], N. COMES[1], D. SOTO[1,2], A. GUAL[1,2], J. P. GIBLIN[1], C. ROZA[3], *X. GASULL[1,2]
[1]Univ. De Barcelona, Barcelona, Spain; [2]IDIBAPS, Barcelona, Spain; [3]Dpto. Biología de Sistemas, Univ. de Alcala, Alcala de Henares, Madrid, Spain

Abstract: Pyrethroid insecticides such as permethrin, deltamethrin or tetramethrin are widely used for pest control, in agriculture and in human public health commonly as a topical treatment for scabies and head lice. Exposure to pyrethroids causes sensory alterations such as transient pain, burning, stinging sensations and paresthesias. Despite their well-known effects on sodium channels, their toxic effects on mammals can’t be fully explained by the modulation of these channels and the contribution of other channels that control sensory neuron excitability is less

studied. Given the role of two-pore domain potassium (K_{2P}) channels in modulating sensory neuron excitability and firing, both in physiological and pathological conditions, we pursued the possibility that pyrethroids modulate K_{2P} channels mainly expressed in sensory neurons. By combining electrophysiological and calcium imaging experiments, we show that a high percentage of tetramethrin-activated neurons were nociceptors, which were also activated by TRPA1 and/or TRPV1 agonists. This pyrethroid produced a significant inhibition of native TRESK, TRAAK, TREK-1 and TREK-2 currents from sensory neurons. Similar effects were found in transfected HEK293 cells expressing the different channels and challenged with pyrethroids. In addition, tetramethrin activated and enhanced the excitability of peripheral saphenous nerve fibers. At the behavioral level, intradermal tetramethrin injection in the mouse paw produced abundant nocifensive responses and caused mechanical allodynia, demonstrating that the effects seen on nociceptors in culture lead to pain-associated behaviors *in vivo*. In TRESK knockout mice, pain-associated behaviors elicited by tetramethrin were enhanced, providing further evidence for a role of this channel in preventing excessive neuronal activation. Our results indicate that inhibition of K_{2P} channels facilitates sensory neuron activation and increases their excitability. These effects contribute to the generation of paresthesias and pain after pyrethroid exposure and point out K_{2P} channels as possible targets of algesic substances.

Disclosures: **A. Castellanos:** None. **A. Andres:** None. **L. Bernal:** None. **G. Callejo:** None. **N. Comes:** None. **D. Soto:** None. **A. Gual:** None. **J.P. Giblin:** None. **C. Roza:** None. **X. Gasull:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.09/EE24

Topic: D.03. Somatosensation: Pain

Title: A microneurography inter-species study comparing the relative distribution of C-nociceptor types and parameters of activity-dependent slowing (ADS) of conduction velocity reflecting different axonal membrane properties in mice, rat, non-human primate and healthy subjects

Authors: *E. GARCIA-PEREZ[1], P. S. PALL[2], R. SOLA[1], M. SUMALLA[1], A. HOUGHTON[2], J. SERRA[1]
[1]Neurosci. Technologies, Barcelona, Spain; [2]Pharmacol., Merck Res. Labs., West Point, PA

Abstract: Objectives: To compare the different proportions of peripheral C-nociceptors in different mammalian species commonly used in preclinical drug development for neuropathic pain in humans.
Methods: Microneurographic recordings were obtained from the common sciatic nerve trunk of

naïve (Mus) mice (n=8), Sprague-Dawley rats (n=8) and from the superficial peroneal nerve of rhesus monkeys (n=8) and healthy human volunteers (n=24) using insulated tungsten microelectrodes. Relative proportions of C-nociceptors and several measures of ADS were compared between species.

Results: Total number of C-fibers recorded in mice, rats, monkeys and humans were 70, 132, 92 and 242, respectively. Relative proportions of C-nociceptors among all recorded C-fibers were 69% in mice, 71% in rats, 47% in monkeys and 79% in humans. Among C-nociceptors, the relative distribution between Type 1A mechano-sensitive and Type 1B mechano-insensitive nociceptors was mice (T1A 58%, T1B 19%), rats (T1A 46%, T1B 47%), monkeys (T1A 40%, T1B 40%) and human (T1A 47%, T1B 43%). A 15±8% of the nociceptors had features that prevented a clear-cut classification to one group or the other and remained as unclassified type C-nociceptors. Baseline conduction velocity (m/s) was: mice (T1A 0.70±0.02, T1B 0.88±0.09), rats (T1A 0.74± 0.03, T1B 0.68± 0.03), monkeys (T1A 0.83±0.03,T1B 0.86±0.16) and humans (T1A 0.73±0.03, T1B 0.48±0.04). Unpaired t-test was statistically significant for mice vs monkeys ($p=0.0041$) for type 1A fibers and for mice vs rats ($p=0.007$) and for mice vs rats ($p=0.007$), mice vs humans ($p=0.0005$), rats vs humans ($p=0.0002$) and monkeys vs humans ($p=0.0005$) for type 1B nociceptors. Percentage of CV slowing at 0.25Hz stimulation was: mice (T1A 0.004±0.15%, T1B 2.93±0.32%), rats (T1A 0.58±0.09%, T1B 3.4±0.25%), monkeys (data unavailable) and human (T1A 0.42±0.18%, T1B 3.87±0.21%). Differences for type 1A fibers were found for mice vs rats ($p=0.0006$) and mice vs humans ($p=0.0021$). Percentage of CV slowing after a 3 min 2Hz stimulation tetanus was mice (T1A 29.71±1.7%, T1B 31.41±3.8%), rats (T1A 31.3±1.58%, T1B 38.5±1.34%), monkeys (T1A 27.39±1.68, T1B 49.6±2.03) and humans (T1A 30.74±1.48%, T1B 34.56±1.9%). Differences were found between mice and rats ($p=0.04$), mice vs monkeys ($p<0.0001$) and monkeys vs humans ($p=0.0005$) only for type 1B fibers.

Conclusions: Although there are minor differences, peripheral C-nociceptor types exhibit remarkable similarity between species in terms of distribution of CV and different measures of ADS. The information obtained in this study is highly relevant when planning preclinical in-vivo studies for peripherally-acting neuropathic pain analgesics.

Disclosures: **E. Garcia-Perez:** A. Employment/Salary (full or part-time):; NEUROSCIENCE TECHNOLOGIES. **P.S. Pall:** A. Employment/Salary (full or part-time):; Merck Research Laboratories. **R. Sola:** A. Employment/Salary (full or part-time):; NEUROSCIENCE TECHNOLOGIES. **M. Sumalla:** A. Employment/Salary (full or part-time):; NEUROSCIENCE TECHNOLOGIES. **A. Houghton:** A. Employment/Salary (full or part-time):; Merck Research Laboratories. **J. Serra:** A. Employment/Salary (full or part-time):; NEUROSCIENCE TECHNOLOGIES.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.10/EE25

Topic: D.03. Somatosensation: Pain

Title: Effect of eugenol on slow ventral root potentials in neonatal rats

Authors: *S. YAGURA, H. ONIMARU, M. IZUMIZAKI
Dept. of Physiol., Showa Univ. Sch. of Med., Tokyo, Japan

Abstract: Eugenol is contained in several plants including clove and is used as an analgesic drug. In peripheral and central nervous system, this compound modulates neuronal activity through action on voltage-gated ionic channels and on transient receptor potential channels. It has been established that slow ventral root potential induced by ipsilateral dorsal root stimulation in the isolated (typically lumbar) spinal cord of newborn rats reflects the nociceptive response. This *in vitro* experimental model is useful for assessing the actions of analgesics. However, there is no report regarding the effects of eugenol in the *in vitro* experimental model. Therefore, we examined the effects of extracellularly applied eugenol on the spinal reflex response in spinal cord preparations from newborn rats. Spinal cord preparations (Th10 - L5) were isolated from Wistar rats (postnatal day 0-3) under deep isoflurane anesthesia. Preparations were superfused continuously at 2.5-3 ml/min in a 2 ml chamber with artificial cerebrospinal fluid (ACSF) composed of (in mM) 124 NaCl, 5.0 KCl, 1.2 KH_2PO_4, 2.4 $CaCl_2$, 1.3 $MgCl_2$, 26 $NaHCO_3$, and 30 glucose and equilibrated with 95% O_2 and 5% CO_2 at a pH of 7.4. The preparations were maintained at a temperature of 25-26°C. To evaluate the effects of eugenol on putative nociceptive responses, the ipsilateral dorsal root of L5 was stimulated using a glass suction electrode, and the induced reflex response was recorded from the L5 and Th12 ventral roots through a 0.5 Hz high-pass filter. The dorsal root was stimulated every 60 s with a 5-20 V, 200 μs square pulse. We measured the peak amplitude and area of the slow ventral root potentials. The preparations were superfused with ACSF for at least 15-30 min until the spinal reflex response became stable. We found that eugenol (0.25-1 mM) caused dose-dependent attenuation of the reflex response. Eugenol also depressed spontaneous ventral root activity. Our report further provides the basic neuronal mechanisms to support the clinical use of eugenol that seems to be potentially expected for analgesic treatment.

Disclosures: **S. Yagura:** None. **H. Onimaru:** None. **M. Izumizaki:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.11/EE26

Topic: D.03. Somatosensation: Pain

Title: Sympathetic modulation of TNFalpha-induced nociception in the presence of oral squamous cell carcinoma

Authors: *N. SCHEFF[1], A. K. SHARMA[2], E. DOWSE[2], B. L. SCHMIDT[3]
[2]Col. of Dent., [3]Bluestone Ctr. for Clin. Res., [1]New York Univ., New York, NY

Abstract: Oral cancer patients report severe pain during function. Oral cancer pain is hypothesized to be due to tumor proliferation, perineural invasion, secretion of pro-nociceptive mediators, and infiltration of immune cells into the cancer microenvironment. Norepinephrine (NE), a key neurotransmitter released by sympathetic nerve fibers, is thought to modulate the immune system and pain in the presence of injury. NE also dysregulates cytokines and growth factors involved in cancer progression. We recently demonstrated that the pro-inflammatory cytokine, tumor necrosis factor alpha (TNFα), mediates oral cancer-associated inflammation and nociception. We sought to test the hypothesis that NE increases TNFα secreted by oral cancer. Using a retrograde tracer, we identified sympathetic postganglionic neurons in the superior cervical ganglia that innervate the tongue. We also showed that TNFα receptors, TNFR1 and TNFR2, are expressed on trigeminal primary afferent neurons innervating the tongue. Membrane β2-adrenergic receptors were expressed on oral cancer cell lines (SCC4, HSC3) and a dysplastic oral keratinocyte cell line (DOK). We measured NE-induced changes in TNFα expression in the cell lines and TNFα protein concentration in the supernatant of each cell line. A 24-hour exposure to 1μM NE did not change TNFα expression or protein concentration. Treatment with 10μM NE, however, elevated TNFα mRNA expression in SCC4 and HSC3 (1.5-fold and 2.3-fold, respectively) and resulted in an increase in TNFα protein in the cell line supernatants of SCC4 and HSC3 (130.9 ± 25.3 and 142.2 ± 12.5% respectively). Furthermore, 10μM NE treatment did not affect mRNA expression in DOK and only resulted in 33.8 ± 1.9% increase in TNFα protein in the DOK supernatant. Pre-treatment with the nonspecific β antagonist, propranolol, blocked the NE-induced increase in TNFα in all cell lines. Together these data suggest that sympathetic neurons in the oral cancer microenvironment potentially release NE that activates oral cancer cells to increase secretion of TNFα. The NE-induced TNFα might activate associated receptors on nociceptive neurons that innervate the cancer microenvironment and cause pain. Ongoing studies in oral cancer animal models are testing the therapeutic effect of β antagonists on cancer pain and inflammation through disruption of TNFα signaling.

Disclosures: **N. Scheff:** None. **A.K. Sharma:** None. **E. Dowse:** None. **B.L. Schmidt:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.12/EE27

Topic: D.03. Somatosensation: Pain

Support: NIH Grant NS065926

NIH Grant NS098826

Title: Translational profiling reveals widespread changes in translation resulting from chemotherapy induced painful neuropathy

Authors: *A. WANGZHOU[1], S. MEGAT[2], T.-F. LOU[3], P. BARRAGAN-IGLESIAS[4], J. K. MOY[4], M. N. ASIEDU[4], P. R. RAY[1], M. D. BURTON[4], G. DUSSOR[4], Z. CAMPBELL[3], T. J. PRICE[4]
[1]Behavioral and Brain Sci., Univ. of Texas At Dallas, Richardson, TX; [2]Behavioral and Brain Sci., [3]Biol., [4]Sch. of Behavioral and Brain Sci., Univ. of Texas at Dallas, Richardson, TX

Abstract: Next-generation sequencing technologies provide unbiased, high throughput snapshots of genomic, transcriptional and nascent translational profiles in cells or tissues. RNA-seq has been used to uncover transcriptional profiles underlying pathologies that could potentially lead to identification of therapeutic targets. However, transcriptomic-based approaches fail to accurately capture how disease states influence the landscape of nascent protein synthesis. We overcome this problem by coupling RNA-seq with Translating Ribosome Affinity Purification (TRAP) technology. We coupled RNA-seq to TRAP (eGFP-tagged ribosomal protein L10a (Rpl10a)) methodology, driven by the nociceptor-specific promoter of Scn10a (Nav1.8), to evaluate mRNA translational efficiency in the mouse dorsal root ganglia (mDRG). We used paclitaxel-induced neuropathic pain model to elucidate changes to the nascent translatome in mDRG neurons for chemotherapy-induced painful neuropathy (CIPN). The Scn10a promoter-driven eGFP-RPl10a was specifically expressed in nociceptors as indicated by co-staining with Prph and CGRP while only a small fraction of large diameter neurons expressed eGFP. Immunoprecipitation (IP) of eGFP from mDRG extracts successfully recovered polyribosome-associated mRNAs from the eGFP-Rpl10a reporter line, with little RNA recovered from mice without the transgene. mRNAs purified by eGFP IP from sensory neurons remained intact and were enriched for nociceptor-specific genes, including Scn10a and Calca/Calcb(CGRP). Traditional transcriptome analysis in the mDRG revealed small differences between naive and CIPN mice, with approximately 200 and 60 genes being up or down regulated, respectively, under paclitaxel treatment compared to naive mice. However, drastic changes were observed in nascent translatome with about 1000 and 900 genes being up or down regulated respectively, indicating pervasive changes in gene expression at the nascent

translatome level. Gene families of Cacn, Calc, Camk, Cox, Eif, Kcn, Ngf, and P2r (among others) are highly translated in CIPN. TRAP-based sequencing provides a novel method for discovering nociceptor-specific mRNAs that are translated in the setting of neuropathic pain. Our CIPN findings clearly demonstrate genome wide changes in translation that cannot be accounted for by transcriptional changes alone. We propose that transcriptional and translational changes are mostly decoupled in nociceptor plasticity, promoting neuropathic pain.

Disclosures: A. Wangzhou: None. **S. Megat:** None. **T. Lou:** None. **P. Barragan-Iglesias:** None. **J.K. Moy:** None. **M.N. Asiedu:** None. **P.R. Ray:** None. **M.D. Burton:** None. **G. Dussor:** None. **Z. Campbell:** None. **T.J. Price:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.13/EE28

Topic: D.03. Somatosensation: Pain

Support: NIH RO1 NS87988

NIH RO1 DE17794

NIH RO1 DE22743

Title: PD-L1 inhibits acute and chronic pain by suppressing nociceptive neuron activity via PD-1

Authors: *Y. KIM[1,2], G. CHEN[1,3], H. LUO[1], D. LIU[1], Z. ZHANG[1], M. LAY[1], W. CHANG[1], R.-R. JI[1]
[1]Dept. of Anesthesiol. and Neurobio., Duke Univ. Med. Ctr., Durham, NC; [2]Dept. of Physiology, Col. of Med., Gachon Univ., Incheon, Korea, Republic of; [3]Nantong Univ., Nantong, China

Abstract: Programmed cell death ligand-1 (PD-L1) is typically produced by cancer cells and suppresses immunity through the receptor PD-1 expressed on T cells. However, the role of PD-L1 and PD-1 in regulating pain and neuronal function is unclear. Here we report that both melanoma and normal neural tissues including dorsal root ganglion (DRG) produce PD-L1 that can potently inhibit acute and chronic pain. Intraplantar injection of PD-L1 evoked analgesia in naive mice via PD-1, whereas PD-L1 neutralization or PD-1 blockade induced mechanical allodynia. Mice lacking *Pd1* (*Pdcd1*) exhibited thermal and mechanical hypersensitivity. PD-1 activation in DRG nociceptive neurons by PD-L1 induced phosphorylation of the tyrosine phosphatase SHP-1, inhibited sodium channels and caused hyperpolarization through activation of TREK2 K^+ channels. PD-L1 also potently suppressed nociceptive neuron excitability in human DRGs. Notably, blocking PD-L1 or PD-1 elicited spontaneous pain and allodynia in

melanoma-bearing mice. Our findings identify a previously unrecognized role of PD-L1 as an endogenous pain inhibitor and a neuromodulator.

Disclosures: Y. Kim: None. **G. Chen:** None. **H. Luo:** None. **D. Liu:** None. **Z. Zhang:** None. **M. Lay:** None. **W. Chang:** None. **R. Ji:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.14/EE29

Topic: D.03. Somatosensation: Pain

Title: Visualization of P2Y1 reporters in peripheral tissues and sensory neurons and changes in response to injury

Authors: C. ESANCY, S. SANNAJUST, T. E. KING, *D. C. MOLLIVER
Biomed. Sci., Univ. of New England, Biddeford, ME

Abstract: P2Y1 is a Gq-coupled receptor for extracellular adenine nucleotides, with highest affinity for adenosine diphosphate (ADP). Functional effects of P2Y1 have been reported in diverse tissues. In peripheral sensory neurons of the dorsal root ganglion (DRG), P2Y1 is one of the most highly expressed P2Y family members and has been implicated in mechanical transduction. P2Y1 null mutant mice show deficits in heat and cold transduction, but the broad non-neuronal expression of P2Y1 poses a challenge for interpretation of behavioral experiments in these animals. Furthermore, it has been difficult to characterize the distribution of P2Y1 in functionally defined subsets of DRG neurons due to a lack of antibodies validated for immunohistochemistry. Here, we used mice with an IRES-Cre recombinase cassette downstream of the P2Y1 stop codon to generate 2 lines of P2Y1 reporter mice: a line expressing td-tomato in all CRE-expressing tissues, to visualize expression in non-neuronal cells of the DRG and peripheral tissues, and a neuron-specific line expressing myristylated GFP to visualize expression in DRG neuron subtypes, as well as their axon projections in peripheral tissues and spinal cord. The results were compared to the distribution of P2Y1 mRNA expression in published RNA-seq datasets. Retrogradely labeled DRG neurons were examined for changes in P2Y1 distribution evoked by hindpaw inflammatory insult or nerve injury. Tissue-specific expression was also examined in skin, colon, bladder and cornea. Finally, behavioral approaches to analyzing function of low threshold afferents expressing P2Y1 were explored in wild type and P2Y1-/- mice. Preliminary results suggest the intriguing possibility that complex social behaviors might be altered in response to deficits in peripheral sensory transduction. However, critical assessment of the challenges inherent in devising behavioral assays for low threshold sensory processing is essential to rigorous interpretation of the results and will be addressed.

Disclosures: **C. Esancy:** None. **S. Sannajust:** None. **T.E. King:** None. **D.C. Molliver:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.15/FF1

Topic: D.03. Somatosensation: Pain

Support: NIH Grant AR057194

Title: Serotonin receptor subtypes responsible for calcium influx in primary cultures from rat dorsal root and trigeminal ganglia

Authors: *D. T. DOMOCOS[1,2], T. SELESCU[1], E. CARSTENS[2], M. IODI CARSTENS[2], A. BABES[1]
[1]Fac. of Biology, Univ. of Bucharest, Bucharest, Romania; [2]Neurobiology, Physiol. and Behavior, Univ. of California, Davis, Davis, CA

Abstract: Serotonin (5-HT) is known to mediate both pain and itch in the peripheral nervous system. Our aim was to identify the specific 5-HT receptor subtypes expressed in the rat dorsal root (DRG) and trigeminal (TG) ganglion neurons.
We used calcium microfluorimetry, the patch clamp technique and pharmacological tools to investigate the action of 5-HT in primary cultures of rat sensory neurons.
Dorsal root and trigeminal ganglia were dissected out from adult male Wistar rats and dissociated neurons were cultured on glass coverslips. After 24 h, the cells were loaded with Calcium Green-1 AM and imaged while being chemically stimulated using a fast-exchange superfusion system.
The calcium responses to 5-HT (50 μM) were classified as transient and sustained according to their kinetics. The selective 5-HT3 antagonist granisetron (1 μM) completely inhibited the transient responses while the 5-HT3 agonist SR 57227 (1 – 10 μM) elicited similar responses in the same neurons.
The non-selective agonist 5-Carboxamidotryptamine (5-CT, 50 μM) induced only sustained responses, solely in the neurons that were sensitive to 5-HT. The same kinds of responses were elicited by the 5-HT1A and 5-HT7 agonist 8-OH-DPAT (10 μM) but were not inhibited by the 5-HT5A antagonist SB-699551-A (1 – 10 μM) or the 5-HT7 antagonist SB269970 (1 – 10 μM).
The 5-HT1A receptor antagonist WAY-100,635 (100 nM) was able to irreversibly inhibit the sustained responses evoked by 5-HT.
Given that the 5-HT1A is metabotropic receptor, we investigated the intracellular mechanisms that led to the sustained calcium influx. The broad-spectrum Transient Receptor Potential (TRP) channel blocker Ruthenium Red did not inhibit the sustained responses. Nor did the more selective antagonists: CIM 0216 (1 μM), A967079 (5 – 10 μM) and HC-067047 (0.5 μM) for

TRPM3, TRPA1 and TRPV4 respectively.
The adenylyl cyclase activator forskolin (10 μM) or the cAMP analog, 8-Br-cAMP (10 μM) did not affect the sustained responses to 5-HT.
Recently reported activators or inhibitors of 5-HT1F, 5-HT2 and 5-HT7 receptors from mouse DRG neurons, had no effects on rat neurons. Voltage clamp experiments were carried out in 5-HT-sensitive DRG neurons pre-selected with calcium imaging, and revealed only transient currents (5-HT3-like) at a holding potential of -80 mV. These currents were inhibited by granisetron (1 μM).
In conclusion, our results suggest that the transient responses are mediated by the 5-HT3 ion channel while the sustained responses are likely mediated by the 5-HT1A metabotropic receptor. This is supported by the inhibitory effect of WAY-100,635 and by the calcium influx elicited by 8-OH-DPAT and 5-CT.

Disclosures: D.T. Domocos: None. **T. Selescu:** None. **E. Carstens:** None. **M. Iodi Carstens:** None. **A. Babes:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.16/FF2

Topic: D.03. Somatosensation: Pain

Title: Sex differences in axonal membrane properties of peripheral C-nociceptors in humans and rats detected by microneurography

Authors: *R. SOLA, M. SUMALLA, J. SERRA
Neurosci. Technologies SLP, Barcelona, Spain

Abstract: Background: Pain perception studies have shown that healthy women perceive noxious stimuli as more painful than men. Furthermore female rodents are also more sensitive than males to noxious stimuli. Nevertheless there are no consistent electrophysiological studies showing such differences in the physiology of peripheral nociceptors.
Objective: To study if there exist any basic axonal membrane property difference between sexes in human and rodent peripheral C-nociceptors.
Methods: Microneurographic recordings of C-fibers were obtained from the peroneal superficial nerve of 22 healthy subjects, 12 females (38.9±3.3 yo) and 10 males (37.7±3.7 yo) and from the sciatic nerve of 52 naïve Sprague-Dawley rats, 25 females and 27 males We analyzed different parameters of activity-depending slowing of conduction velocity (ADS) and conduction velocity.
Results: A total of 141 C-nociceptors (75 mechano-heat-responsive [CMH] and 66 mechano-insensitive afferents [MIA])
among all measured ADS parameters only but striking difference in C fibers conduction velocity

has been identified. There is a statistically significant higher CV for human male's C fibers (0,70 ± 0,18) than for female's C fibers (0,51 ± 0,25), (P<0.001), as well for CMH (P<0.024) but no for MIA afferents (p<0.06). In rats, , have been studied in a similar manner recording a total of 87 C nociceptors (42 CMH and 45 MIA). CV of CMH units is higher as well (p<0.046). It is important to highlight a clear different C fiber CV distribution, being the coefficient of variation much higher for human females (45.4%) than for males (15.5%)
Discussion: this study has shown different CV in C-nociceptors, especially in CMH units, according to sex both in humans and rats. CMH units are responsive to mechanical and heat noxious stimuli. Our study suggests that the combination of lower CV and higher range of CV distribution for CMH units could modulate the temporal code of peripheral noxious input in such a way that results in increased pain perception in females and thus could partly explain gender differences in pain perception.
Conclusion: Peripheral C-nociceptor conduction velocity is lower in females than in males. This electrophysiological feature could explain differences in pain perception between sexes in humans and rodents.
fig 1. significant higher CV for human male's C fibers (0,70 ± 0,18) than for female's C fibers (0,51 ± 0,25), (P<0.001), as well for CMH (P<0.024)
Table 1

Disclosures: **R. Sola:** A. Employment/Salary (full or part-time):; full-time employee at NT. **M. Sumalla:** A. Employment/Salary (full or part-time):; full-time employee at NT. **J. Serra:** A. Employment/Salary (full or part-time):; full-time employee at NT.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.17/FF3

Topic: D.03. Somatosensation: Pain

Title: Assessment of Nav1.7-dependent electrical excitability of DRG neurons using a high-capacity calcium influx assay

Authors: L. DENG[1], K. STARK[1], S. LARDELL[2], P. KARILA[2], *D. H. HACKOS[1]
[1]Neurosci., Genentech Inc, South San Francisco, CA; [2]Cellectricon AB, Molndal, Sweden

Abstract: The sodium channel Nav1.7 is an attractive pain target based on human genetic evidence. Gain-of-function mutations in Nav1.7 are associated with inherited severe pain conditions, such as inherited erythromelalgia (IEM) or paroxysmal extreme pain disorder (PEPD), whereas individuals carrying loss-of-function mutations are congenitally insensitive to pain (CIP). Voltage-clamp recordings reveal that Nav1.7 makes up >80% of TTX-sensitive Nav current in small-diameter DRG neurons in culture. Current clamp recordings further demonstrate

that action potential generation in small-diameter DRG neurons is dependent on Nav1.7 as long as the resting membrane potential is adjusted to -70mV or more negative (where Nav1.7 is not fully inactivated). However, such electrophysiological recordings are time-consuming not well suited for testing the effects of Nav1.7-selective inhibitors. Here we demonstrate a medium-throughput assay that allows measurements of calcium influx following field electrical stimulation of DRG neurons in culture. By comparing WT vs Nav1.7 KO DRG neurons, we find that Nav1.7 is absolutely required for the observed electrical excitability and calcium influx. Furthermore, we demonstrate the use of high-content imaging to examine the excitability of individual subtypes of DRG neurons and their dependence on Nav1.7. Finally, we examine the effects of Nav1.7-selective inhibitors on DRG excitability.

Disclosures: **L. Deng:** A. Employment/Salary (full or part-time):; Genentech, Inc. **K. Stark:** A. Employment/Salary (full or part-time):; Genentech, Inc. **S. Lardell:** A. Employment/Salary (full or part-time):; Cellectricon AB. **P. Karila:** A. Employment/Salary (full or part-time):; Cellectricon AB. **D.H. Hackos:** A. Employment/Salary (full or part-time):; Genentech, Inc..

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.18/FF4

Topic: D.03. Somatosensation: Pain

Title: Role of nociceptors in driving B cell antibody class switching in allergic inflammation

Authors: *S. MATHUR[1,2], S. FOSTER[2], C. SEEHUS[2], S. TALBOT[2], C. J. WOOLF[2]
[1]Harvard Univ., Cambridge, MA; [2]Neurobio., Children's Hosp. Boston, Boston, MA

Abstract: During allergic inflammation, B-cells secrete antibodies that induce deleterious immune responses, including recruitment of immune cells to the site of the allergen exposure and smooth muscle contraction, potentially with deadly consequences in the airway in particular. In response to allergens, antibody genes re-arrange in a highly coordinated manner within B cells, a phenomenon termed immunoglobin class switching, allowing B cells to produce antibodies of different subtypes to modulate the immune response. The antibody IgE is released in an allergic response and directly activates mast cells, which then cause the release of inflammatory mediators, which can initiate bronchoconstriction, hyperperistalsis, and sensitize the pain pathway. Recent evidence has shown that nociceptors, high-threshold sensory neurons that detect noxious stimuli and elicit the sensation of pain, interact with immune cells to modulate their response during inflammatory disease. However, little is known about the interaction of B cells with nociceptors. We previously found that in animal models of allergic airway inflammation, genetic ablation and pharmacologic silencing of nociceptors results in decreased T helper type-2 cell (Th2) effector cytokine function, which potentially could impact the recruitment and

activation of B-cells in germinal centers. Here, we wanted to determine the role of nociceptors if any in modulating B cell class switching to IgE. Interestingly, we found that in the absence of nociceptors, mice sensitized to two different allergens had significantly less serum IgE than wild-type (WT) sensitized mice. In addition, we found that *in vitro* stimulation of B cells from nociceptor ablated mice produced similar levels of serum IgE as WT, implying that the decrease in IgE may not be an intrinsic developmental defect in B cells. Furthermore, co-culture of WT B cells with nociceptors in the absence of additional stimulation led to production of IgE. Our results suggest that nociceptors produce molecules that regulate the B cell class switch pathway. Studying these interactions may provide novel treatment strategies for allergic asthma and other debilitating autoinflammatory diseases.

Disclosures: S. Mathur: None. **S. Foster:** None. **C. Seehus:** None. **S. Talbot:** None. **C.J. Woolf:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.19/FF5

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: NIGMS Grant P20GM103643

Title: Characterization of the loss of primary cilia in nociceptive dorsal root ganglion neurons and its affect on acute pain processing

Authors: *K. L. LINDROS[1], E. J. BILSKY[3], K. L. TUCKER[2]
[2]Dept. of Biomed. Sci., [1]Univ. of New England, Biddeford, ME; [3]Admin., Pacific Northwest Univ. of Hlth. Sci., Yakima, WA

Abstract: In sensory DRG neurons, robust primary cilia appear at the soma shortly after elaboration of axons in embryonic development as early as embryonic day E12.5 and persist throughout postnatal stages of maturation. Primary cilia can be functionally eliminated through inducible inactivation of genes that encode proteins essential for the maintenance of primary cilia, such as the intraflagellar transport protein *Ift88.* To this end, knocking out the maintenance gene in the specific area of interest, nociceptive neurons in DRGs, constructs a working model for lack of cilia in sensory areas that largely control pain processing. We have chosen to focus on eliminating cilia from two different nociceptive neuronal subtypes, using an inducible knockout allele of *Ift88* crossed to transgenic mice expressing Cre-recombinase under the control of either the TrpV1 or $Na_V1.8$ promoter. Examination of E15.5 embryos that are Cre-recombinase positive and homozygous for the *Ift88* floxed allele revealed a loss of primary cilia in DRG neurons. This was made easier by the introduction of the lineage tracker Ai14 to the Cre driver, marking all

Cre-expressing TrpV1 neurons with a fluorescent tdTomato reporter. Subsequently, a variety of standard behavioral phenotyping including locomotor activity assays, gait and performance assays, as well as thermal and mechanical withdrawal assays, were conducted. Nociceptive testing assays following injection of Complete Freud's Adjuvant and Capsaicin were also completed for indications of initial deficits in sensory/pain processing. Finally, exploration of the role that primary cilia play in the development and maintenance of nociceptors was carried out to develop a hypothesis for the potential clinical applications of primary cilia in acute and chronic pain states.

Disclosures: K.L. Lindros: None. **E.J. Bilsky:** None. **K.L. Tucker:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.20/FF6

Topic: D.03. Somatosensation: Pain

Support: NIH R01 NS065926

NIH R56 NS098826

Title: The evolutionary history of human dorsal root ganglia - enriched genes: Regulatory and coding sequence evolution as windows into functional turnover

Authors: *P. R. RAY[1], ***P. R. RAY**[1], T. J. PRICE[2]
[1]Behavioral and Brain Sci., The Univ. of Texas at Dallas, Richardson, TX; [2]Behavioral and Brain Sci., UTD, Richardson, TX

Abstract: Problem: Human dorsal root ganglia (DRG) and trigeminal ganglia are known to express many tissue-restricted genes, although the evolutionary history of these genes have not been well characterized except in the context of specific species like the vampire bat or star-nosed mole. As a result, it is difficult to predict if the molecular and functional roles of gene products are altered due to positive selection in the coding sequence, or whether programs controlling transcription or translation have changed due to regulatory evolution. We analyze evolutionary conservation of gene expression patterns and sequence to analyze this. **Approach and Results:** We identify DRG-enriched genes with potentially conserved transcription regulatory programs between humans and mouse by finding conserved tissue specificity of gene expression, avoiding explicit evolutionary analysis of regulatory sequences due to challenges of identifying distal regulatory regions. The set of genes with conserved DRG-enriched expression across species include many well-known DRG-related genes (PRDM12, TRPV1, SCN10A) but also have several notable omissions (TRPM8, TRPA1) which show regulatory divergence.

Analysis of gene families in the set of genes with conserved DRG enrichment shows two main groups of genes with Most Recent Common Ancestors that can be traced back to Vertebrata or Bilateria clades, suggesting that some of these genes co-evolved with nerve chords or dorsal root ganglia in animals. Identification of gene families with large numbers of duplication events show a signature of duplication followed by diversification of gene expression [Lynch and Conery, Science 2000], like members of the MRGPR family. Analysis of gene sequences yields several genes with evidence of recent selection in human populations, including glutamate receptors like GRIA2 and GRIK1 (with weaker enrichment but known expression and functional roles in DRG) [Schrider and Kern, Biorxiv 2017]. **Discussion:** Unlike olfactory, taste or vision receptors, somatosensory genes are less prone to adaptive sequence evolution, though a subset of them show divergent expression between human and mouse, suggesting turnover of regulatory function. Comprehensive analysis can reveal a blueprint of mammalian evolution for genes related to somatosensation, similar to in-depth evolutionary studies performed on olfactory receptors [Niimura and Nei, PNAS 2003]. Additionally, high rates of translational failure in preclinical therapeutic models [Van der Worp *et al*, PLoS Medicine 2014] could potentially be lowered by informed choice of both drug targets (gene products), and model systems based on evolutionary analysis.

Disclosures: P.R. Ray: None. **T.J. Price:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.21/FF7

Topic: D.03. Somatosensation: Pain

Support: NIH Grant NS065926

NIH Grant NS098826

Title: Genome-wide nonparametric tests of exon expression identify differentially expressed transcripts in human dorsal root ganglia and tibial nerves with sexual dimorphism

Authors: *J. KHAN[1], C. RAO[2], T. J. PRICE[3], P. R. RAY[3]
[1]Sch. of Natural Sci. and Mathematics, Univ. of Texas At Dallas, Richardson, TX; [2]Erik Jonsson Sch. of Engin. and Computer Sci., Univ. of Texas at Dallas, Richardson, TX; [3]Sch. of Behavioral and Brain Sci., UTD, Richardson, TX

Abstract: Problem: Tests for differential gene expression in *in vivo*, high throughput transcriptome profiling assays for human datasets typically rely on parametric statistics, due to small sample sizes resulting from regulatory and financial constraints in sourcing tissue from

large donor cohorts. Recently, the GTEx consortium [GTEx *et al*, Science 2015] and several laboratories have performed RNA-seq on a wide range of human tissues at unprecedented scale, allowing systematic analysis for *in vivo* human gene expression using nonparametric tests [Li and Tibshirani, SMMR 2013]. We developed a tool for performing nonparametric tests on distribution or sample statistics for mRNA expression at the resolution of individual exons in subsets of human transcriptome profiles defined by biological (tissue or cell type, sex, pathology), or clinical variables and used it to investigate sexual dimorphism in the tibial nerve. We sequenced dorsal root ganglia (DRG) from 6 human donors, and integrated our datasets with 304 tibial nerve samples from GTEx for analysis.
Results: We used 199 male and 105 female tibial nerve samples to identify exons or exon-exon junction boundaries that are differentially expressed between males and females in the tibial nerve. After removing sex linked genes, our tool identified genes that are consistently differentially expressed between males and females, including several related to neuronal function or neuropathies. COMP was found to be upregulated 2 fold in males and is part of the thrombospondin family known to be involved in synaptogenesis, although the role of COMP remains to be elucidated. GSTM1 and HSD11B1, both involved in glucocorticoid signaling, were upregulated 2-fold or more in females and are known to play a role in diabetic neuropathy and inflammation response respectively. MMP3 (Matrix Metallopeptidase 3) plays a role in mammalian inflammation models and pain processing, and is significantly increased in males.
Discussion: Multiple molecular mechanisms of pain and pain inhibition are known to possess sex differences, which is often overlooked in preclinical models that control for sex [Mogil, Nature 2016], increasing the potential for translational failure. Thus, our genome wide mRNA profile of sexual dimorphism in human peripheral nerves is relevant from a biological and pharmacogenomics perspective. Our computational framework opens up the possibility of performing differential whole gene, transcription start site or splicing analysis for other sensory tissues like the trigeminal ganglia, while making fewer assumptions about potentially diverse clinical samples due to use of nonparametric tests.

Disclosures: **J. Khan:** None. **C. Rao:** None. **T.J. Price:** None. **P.R. Ray:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.22/FF8

Topic: D.03. Somatosensation: Pain

Support: Zilkha Family Discovery Fellowship in Neuroengineering

NIH (R01 NS091759)

US Army Medical Research (W81XWH-15-1-0702)

Title: Distinct classes of primary nociceptors show electrophysiological specializations for driving ongoing and evoked pain

Authors: *M. A. ODEM, R. M. CASSIDY, A. BAVENCOFFE, E. LOPEZ, J. TIAN, Z. WU, C. W. DESSAUER, E. T. WALTERS
Integrative Biol. and Pharmacol., McGovern Med. Sch. At UTHealth, Houston, TX

Abstract: Persistent ongoing pain, hyperalgesia, and allodynia following spinal cord injury (SCI) in rats requires continuing activity in nociceptors (Yang et al., *J Neurosci*, 34:10765, 2014). For months after SCI, many nociceptors generate spontaneous activity (SA) in their somata, both *in vivo* and after dissociation (Bedi et al., *J Neurosci*, 30:14870, 2010; Wu et al., *Pain*, 154:2130, 2013; Bavencoffe et al., *J Neurosci*, 36:1660, 2016). We have found that small (15-30 µm soma) DRG neurons dissociated from naïve rats and tested 20 h later fall into 2 distinct classes. **RF neurons** show non-accommodating **repetitive firing** during prolonged depolarization. **Ac neurons** show strong **AP accommodation** without repetitive firing at any level of depolarization. Both classes are primarily nociceptors, with ~70% excited by 1 µM capsaicin, and 40-60% also binding isolectin B4. RF neurons have smaller AP amplitudes and slower rise times, as well as longer afterhyperpolarizations that limit firing frequency during ongoing activity. RMP does not differ between the classes. In principle, SA might be generated by 1) tonic depolarization of resting membrane potential (RMP), 2) decreased action potential (AP) threshold, and/or 3) increased amplitude of depolarizing, spontaneous fluctuations (DSFs) of MP. We found that SCI induces chronic SA by all three effects, but only in RF neurons (~60% with SA, versus 0% of Ac neurons). After SCI only RF neurons showed persistently depolarized RMP. AP voltage thresholds are normally lower in RF neurons, and showed further lowering by SCI. Both RF and Ac neurons had lower rheobase after SCI. In A-fibre sensory neurons, sinusoidal, subthreshold oscillations drive SA after nerve injury (Amir et al., *J Neurosci*, 19:8589, 1999). In contrast, RF and Ac neurons generate irregular DSFs that are non-sinusoidal. Large DSFs (5-25 mV), which normally almost never occur in either class, increased dramatically in RF but not Ac neurons after SCI. Large-DSF frequency after SCI was nearly identical to the SA firing rate. Thus, the combination of depolarized RMP, reduced AP threshold, and enhanced DSFs accounts for the chronic generation of SA in RF nociceptors, and likely represents a set of mechanisms specialized to generate irregular, low-frequency firing in RF somata and peripheral terminals that can drive ongoing pain. Ac nociceptors may be specialized for rapid, transient, peripheral activation at the onset of noxious stimuli. Lowering of rheobase in Ac and RF nociceptors, if it also occurs in peripheral terminals, should contribute to hyperalgesia. Supported by Zilkha Family Discovery Fellowship, NIH (R01 NS091759), and US Army Medical Research (W81XWH-15-1-0702).

Disclosures: M.A. Odem: None. **R.M. Cassidy:** None. **A. Bavencoffe:** None. **E. Lopez:** None. **J. Tian:** None. **Z. Wu:** None. **C.W. Dessauer:** None. **E.T. Walters:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.23/FF9

Topic: D.03. Somatosensation: Pain

Support: R01NS100788

R01NS065926

Title: Inhibition of Poly(A)-binding proteins reveals a key role in behavioral responses to inflammatory pain

Authors: *Z. CAMPBELL[1], P. BARRAGAN-IGLESIAS[2], T.-F. LOU[2], B. J. BLACK[5], V. BHAT[2], J. PANCRAZIO[3], T. J. PRICE[4]
[1]Biol., UT Dallas, Richardson, TX; [2]UT-Dallas, Dallas, TX; [4]Sch. of Behavioral and Brain Sci., [3]UT-Dallas, Richardson, TX; [5]The Univ. of Texas At Dallas, Irving, TX

Abstract: Post-transcriptional gene control is a dominant theme in neuronal plasticity. Endogenous cytokines that promote pain plasticity *in vivo* stimulate cap-dependent translation via eukaryotic initiation factor 4 (eIF4) proteins in nociceptors. Far less is known regarding regulatory events that occur on the 3' end. Regulated cytoplasmic polyadenylation serves crucial roles in development and in the nervous system. The direct consequence of poly(A) extension is increased binding of poly(A) binding proteins (PABPs) which stimulate translation initiation through simultaneous associations with the poly(A) tail and eIF4 proteins. PABPs are difficult to study; multiple paralogs produce functional redundancy and there is a paucity of pharmacological reagents. As a novel means of competitively inhibiting PABP function, we conducted an unbiased assessment of PABP specificity. These results were used to develop a specificity derived competitive inhibitor oligonucleotide (SPOT-ON). SPOT-ON RNAs are chemically modified to resist nuclease degradation and display 10 fold greater stability than an unmodified counterpart. The Poly(A) SPOT-ON binds with high affinity and selectivity to PABP. Our inhibitor confirms that PABP functions at the initiation phase of translation. We demonstrate that PABP is required for nascent protein synthesis in primary afferent neurons and their axons. PABP is broadly expressed in soma of cultured DRG sensory neurons and their axons, spinal dorsal horn, the dorsal root ganglia, and sciatic nerve. Mechanical nociceptive plasticity induced by inflammation is blocked by PABP inhibition. Collectively, these results suggest that PABP is integral for nociceptive plasticity.

Disclosures: Z. Campbell: None. **P. Barragan-Iglesias:** None. **T. Lou:** None. **B.J. Black:** None. **V. Bhat:** None. **J. Pancrazio:** None. **T.J. Price:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.24/FF10

Topic: D.03. Somatosensation: Pain

Support: FONDECYT 11121302

FONDECYT 1151043

FONDECYT 1161490

FONDEQUIP EQM140100

FONDECYT 1140325

FONDAP 15150012

Title: Cyclin dependent kinase 5 modulates the P2X2a receptor channel gating through phosphorylation of threonine 372

Authors: *E. UTRERAS PURATICH[1], R. SANDOVAL[1], P. CASTRO[2], P. LAZCANO[1], M. HEVIA[2], M. ROKIC[3], B. HALL[4], A. TERSE[4], S. S. STOJILKOVIC[3], C. GONZALEZ-BILLAULT[1,5], A. B. KULKARNI[4], C. CODDOU[2]
[1]Dept. of Biol., Univ. de Chile, Santiago, Chile; [2]Catholic Univ. of the North, Coquimbo, Chile; [3]Section on Cell. Signaling, NICHD, NIH, Bethesda, MD; [4]Functional Genomics Section, LCDB, NIDCR, NIH, Bethesda, MD; [5]Ctr. for Geroscience, Brain Hlth. and Metabolism (GERO), Santiago, Chile

Abstract: The purinergic P2X2 receptor (P2X2R) is an ATP-gated ion channel widely expressed in the nervous system. Here, we identified a putative Cdk5 phosphorylation site in the full-size variant P2X2aR (^{372}TPKH^{375}), which is absent in the splice variant P2X2bR. We therefore investigated the effects of Cdk5 and its neuronal activator, p35, on P2X2aR function. We found an interaction between P2X2aR and Cdk5/p35 by co-immunofluorescence and co-immunoprecipitation in HEK293 cells. We also found that threonine-phosphorylation was significantly increased in HEK293 cells co-expressing P2X2aR and p35 as compared to cells expressing only P2X2aR. Moreover, P2X2aR-derived peptides encompassing the Cdk5 consensus motif were phosphorylated by Cdk5/p35. Whole-cell patch-clamp recordings indicated a delay in development of use-dependent desensitization (UDD) of P2X2aR but not of P2X2bR in HEK293 cells co-expressing P2X2aR and p35. In *Xenopus* oocytes, P2X2aRs showed a slower UDD than in HEK293 cells and Cdk5-activation prevented this effect. A similar effect was found in P2X2a/3R heteromeric currents in HEK293 cells. The P2X2aR-

T372A mutant was resistant to UDD. In endogenous cells, we observed similar distribution between P2X2R and Cdk5/p35 by co-localization using immunofluorescence in primary culture of nociceptive neurons. Moreover, co-immunoprecipitation experiments showed an interaction between Cdk5 and P2X2R in mouse trigeminal ganglia. Finally, endogenous P2X2aR-mediated currents in PC12 cells, and P2X2/3R mediated increases of intracellular Ca^{2+} in trigeminal neurons were Cdk5-dependent, since inhibition with roscovitine accelerated the desensitization kinetics of these responses. These results indicate that the P2X2aR is a novel target for Cdk5-mediated phosphorylation, which might play important physiological roles including pain signaling.

Disclosures: E. Utreras Puratich: None. **R. Sandoval:** None. **P. Castro:** None. **P. Lazcano:** None. **M. Hevia:** None. **M. rokic:** None. **B. hall:** None. **A. Terse:** None. **S.S. Stojilkovic:** None. **C. Gonzalez-Billault:** None. **A.B. Kulkarni:** None. **C. Coddou:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.25/FF11

Topic: D.03. Somatosensation: Pain

Support: NIH Grant R01NS42595

Title: Intrinsic homeostatic plasticity in mouse and human peripheral nociceptors

Authors: *L. MCILVRIED[1], M. PULLEN-COLON[2], R. W. GEREAU, IV[3]
[1]Anesthesiol., Washington Univ. At St. Louis, Saint Louis, MO; [2]Anesthesiol., Washington Univ. In St Louis, Saint Louis, MO; [3]Anesthesiol., Washington Univ. Sch. Med., Saint Louis, MO

Abstract: The nervous system constantly undergoes experience-dependent plasticity. In response to a change in activity from fluctuations in the environment, homeostatic plasticity permits a compensatory shift of intrinsic excitability, allowing a return to a physiological, basal level. Stabilization of neuronal activity is critical for normal nervous system function, and homeostatic plasticity prevents neural circuits from becoming either hyper- or hypoexcitable. However, if this protective plasticity exists, then how does chronic pain develop? Homeostatic plasticity has been primarily studied in the central nervous system, so to first address this question, we recently tested whether this plasticity also occurs in peripheral nociceptors [i.e., small diameter dorsal root ganglia (DRG) neurons]. We hypothesized that mouse and human primary sensory afferents undergo adaptive changes in neuronal excitability due to intrinsic homeostatic plasticity. Using a combination of pharmacologic and optogenetic approaches, we find that chronic depolarization of mouse DRG neurons leads to a decrease in excitability of

small diameter neurons (putative nociceptors) as shown by decreased input resistance, increased rheobase and decreased number of action potentials in response to suprathreshold current injections. These effects rebounded as an increase in excitability with a 24 hour recovery. Preliminary data indicate that human DRGs show similar responses, and therefore also undergo homeostatic plasticity. Our results show that both mouse and human nociceptors are capable of homeostatic regulation of intrinsic excitability in response to sustained, pharmacological depolarization or sustained stimulation of activity. Primary afferent nociceptors are important for the development of pain, and it will be important to determine whether mechanisms of this plasticity are altered during chronic pain.

Disclosures: L. McIlvried: None. **M. Pullen-Colon:** None. **R.W. Gereau:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.26/FF12

Topic: D.02. Somatosensation

Support: CRACK IT Challenge 9: NC3Rs

Title: Development, validation and functional characterisation of primary cultures of sensory neurons derived from adult human dorsal root ganglia

Authors: G. B. MILES[1], A. K. CHOUHAN[1], A.-C. DEVLIN[1], D. ACTON[1], M. VALDOR[2], S. ESSER-ROSENOW[2], S. FRINGS[2], T. CHRISTOPH[2], A. M. RUSH[3], J. ROBERTS[4], T. REEKIE[4], M. CROUCH[4], *D. I. HUGHES[4], M. O. RIEHLE[4], A. HART[4]
[1]Univ. St Andrews, St Andrews, United Kingdom; [2]Grünenthal GmbH, Aachen, Germany; [3]Metrion Biosci. Ltd., Cambridge, United Kingdom; [4]Univ. of Glasgow, Glasgow, United Kingdom

Abstract: There is a critical need for the establishment, validation and characterisation of human cell-based models in which to study nociceptive processing to aid in the development of novel pain therapeutics. We have therefore developed techniques for the ethical acquisition, culturing and functional analysis of human dorsal root ganglion (DRG) neurons. To date, we have successfully retrieved whole DRGs from organ donors, dissociated and established primary cultures of DRG cells, shipped cultured cells to both national and international consortium member laboratories, and electrophysiologically characterised DRG neurons in culture. Initial immunohistochemical analyses of immersion-fixed free-floating human DRG sections revealed a diversity of sensory neuron subtypes based on expression of NF200, Parvalbumin, CGRP, Somatostatin and substance P, which were each expressed by subsets of cells exhibiting distinct size distributions. Whole-cell patch-clamp recordings (conducted in 3 laboratories; St Andrews,

Metrion and Grunenthal) then demonstrated the functional viability of cultured DRG neurons along with their utility for physiological and pharmacological studies. Current-clamp mode recordings revealed that most DRG neurons generate single (44%) or multiple (28%) action potentials upon injection of square current pulses (n=54 cells). Analysis of action potential parameters revealed average half-widths of 3.6 ± 0.4 ms, and average amplitudes of 58 ± 3.6 mV (n=30 cells). Voltage-clamp mode recordings revealed fast, inactivating sodium currents, that varied in their TTX-sensitivity (21% cells completely TTX-sensitive, 21% completely TTX-resistant, 58% mixed; n=24 cells), transient and persistent voltage-gated potassium currents (23/23 cells), and both high and low voltage-gated calcium currents (10/10 cells). Current responses to the application of specific receptor agonists revealed heterogeneous expression of functional TRPV1 (capsaicin, 27/45 cells), P2X3 receptor (α,β-methyl ATP, 5/16 cells), GABA (GABA, 12/17 cells) and ASICs (low pH, 15/16 cells) receptors in cultured DRG neurons. Taken together, these data demonstrate that a mixed population of sensory neurons, including likely nociceptive afferents, can be successfully cultured from human DRGs and that these cells can be utilised for both electrophysiological and pharmacological studies which will reduce animal use in line with the principles of the 3Rs. This human cell-based model is likely to facilitate advances in our knowledge of the processing of sensory signals by human neurons, and aid in the development and validation of much needed, novel pain therapeutics.

Disclosures: G.B. Miles: None. **A.K. Chouhan:** None. **A. Devlin:** None. **D. Acton:** None. **M. Valdor:** A. Employment/Salary (full or part-time):; Grünenthal GmbH. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Grünenthal GmbH. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Grünenthal GmbH. **S. Esser-Rosenow:** A. Employment/Salary (full or part-time):; Grünenthal GmbH. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Grünenthal GmbH. **S. Frings:** A. Employment/Salary (full or part-time):; Grünenthal GmbH. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Grünenthal GmbH. **T. Christoph:** A. Employment/Salary (full or part-time):; Grünenthal GmbH. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Grünenthal GmbH. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Grünenthal GmbH. **A.M. Rush:** A. Employment/Salary (full or part-time):; Metrion Biosciences Ltd.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Metrion Biosciences Ltd.. **J. Roberts:** None. **T. Reekie:** None. **M. Crouch:** None. **D.I. Hughes:** None. **M.O. Riehle:** None. **A. Hart:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.27/FF13

Topic: D.03. Somatosensation: Pain

Title: Nociceptor sensitivity in *Drosophila* larvae is controlled by RNA-binding proteins that regulate translation

Authors: K. HOFFMAN, R. STEWART, A. DYSON, *A. C. BELLEMER
Biol., Appalachian State Univ., Boone, NC

Abstract: Nociception refers to detection of noxious mechanical, chemical, or thermal stimuli by specialized somatosensory neurons called nociceptors. Sensitization of these nociceptor neurons in response to tissue damage or inflammation is at the root of many chronic pain syndromes. In *Drosophila melanogaster* larvae, the Class IV multidendritic neurons are activated by noxious thermal, mechanical, and UV stimuli to generate a stereotyped behavioral response termed nocifensive escape locomotion. This behavioral response can be investigated using powerful *Drosophila* genetic methods to identify and characterize the cellular and molecular mechanisms that shape nociceptor sensitivity. We have used a nociceptor-specific RNAi screen approach to identify widespread roles for RNA-binding proteins in regulating the sensitivity of nociceptors to noxious thermal and mechanical stimuli. Many of the genes that were found to produce defects in nocifensive escape locomotion when knocked down have been previously characterized as regulators of translation. Specifically, nociceptor-specific knockdown of multiple eukaryotic initiation factor genes caused insensitivity to noxious thermal and mechanical stimuli, while knockdown of the translational repressor gene, *pumilio*, produced hypersensitivity to noxious thermal and mechanical stimuli. These findings indicate that the regulation of protein synthesis is able to bidirectionally regulate the basal sensitivity of *Drosophila* nociceptors and also suggest the hypothesis that protein synthesis may be regulated during the sensitization of nociceptors in response to tissue damage or inflammation. Ongoing experiments will characterize the cellular mechanisms that translational regulators use to shape nociceptor function and also identify the molecular targets subject to translational regulation.

Disclosures: K. Hoffman: None. **R. Stewart:** None. **A. Dyson:** None. **A.C. Bellemer:** None.

Poster

397. Nociceptors

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 397.28/FF14

Topic: D.03. Somatosensation: Pain

Support: NIH 2T32MH067564

Title: A core signaling mechanism at the origin of animal nociception

Authors: E. E. ZAHARIEVA[1], O. M. ARENAS SABOGAL[2], C. VÁSQUEZ-DOORMAN[3], A. PARA[2], C. P. PETERSEN[3], *M. GALLIO[2]
[1]Neurobio., Dept of Neurobio., Evanston, IL; [2]Neurobio., [3]Mol. Biosci., Northwestern Univ., Evanston, IL

Abstract: All animals must detect noxious stimuli to initiate protective behaviors, but the evolutionary origin of nociceptive systems is not well understood. Here, we show that a remarkably conserved signaling mechanism mediates the detection of noxious stimuli in animals as diverse as flatworms and humans. Planarian flatworms are amongst the simplest bilateral animals with a centralized nervous system, and capable of directed behavior. We demonstrate that noxious heat and irritant chemicals elicit robust escape behaviors in the planarian *Schmidtea mediterranea*, and that the conserved ion channel TRPA1 is required in peripheral neurons for these responses. TRPA1 mutant fruit flies (*Drosophila*) are also defective in the avoidance of noxious heat. Unexpectedly, we find that either planarian, or human TRPA1 can restore noxious heat avoidance to TRPA1 mutant *Drosophila*, despite the fact that neither is directly activated by heat - instead, a conserved cellular and molecular mechanism for channel activation is at the core of this remarkable across-phylum rescue. Together, our data reveal a core function for TRPA1 in signal transduction, demonstrate its conservation from planarians to humans, and imply that human nociceptive systems may share a common ancestry with those of most extant animals, tracing back their origin to a progenitor that lived more than 500 million years ago.

Disclosures: E.E. Zaharieva: None. **O.M. Arenas Sabogal:** None. **C. Vásquez-Doorman:** None. **A. Para:** None. **C.P. Petersen:** None. **M. Gallio:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.01/FF15

Topic: D.03. Somatosensation: Pain

Support: ĐFĢ PĿ 321/11-1

ĐFĢ PĿ 321/13-1

Title: The relationship between noxious stimuli, behavioral responses and pain perception in chronic pain patients

Authors: *H. B. HEITMANN[1], E. S. MAY[1], L. TIEMANN[1], P. SCHMIDT[1], S. TA DINH[1], M. M. NICKEL[1], T. R. TÖLLE[2], M. PLONER[1]
[1]Dept. of Neurol. and TUM-Neuroimaging Ctr., [2]Dept. of Neurol., Technische Univ. München, Muenchen, Germany

Abstract: Pain serves vital protective functions, which crucially depend on appropriate behavioral responses to noxious stimuli. However, pain has mostly been conceptualized as a perceptual phenomenon. Recent findings support a more action-oriented concept by showing that behavioral responses to noxious stimuli significantly shape the perception of pain in healthy human subjects. In chronic pain, perception is often decoupled from noxious input and behavioral responses are no longer protective but often unsuccessful or even maladaptive, suggesting fundamental changes of stimulus-perception-behavior relationships in chronic pain. However, direct and quantifiable evidence for these interrelations and their potential changes in chronic pain is lacking. We therefore applied a simple behavioral paradigm to investigate such relationships in 22 chronic pain patients and 22 age-matched healthy control subjects. Patients and controls performed speeded behavioral reactions and provided perceptual ratings of brief noxious and tactile stimuli to their right hand. As expected, intensity ratings increased and reaction times decreased with increasing stimulus intensity for both stimulus types in both patients and controls. In both groups, multi-level moderated mediation analyses revealed a significantly stronger involvement of behavioral responses in the translation of noxious stimuli into perception than in the translation of tactile stimuli. These results confirm previous findings and extend them to an older age group, providing further evidence for an action-oriented concept of pain. Moreover, we found that the mediation effect of behavioral responses in the stimulus-perception relationship was higher in chronic pain patients than in healthy controls, hinting at an even stronger role of behavioral responses for perception in chronic pain. Additional analyses will relate these findings to personality traits and disease-related parameters. This approach promises quantifiable insights into the relationships between noxious stimuli, behavior and pain perception that appear to be fundamentally altered in chronic pain. Eventually, a better

understanding of these complex interrelations might contribute to the development of new treatment strategies in chronic pain.

Disclosures: **H.B. Heitmann:** None. **E.S. May:** None. **L. Tiemann:** None. **P. Schmidt:** None. **S. Ta Dinh:** None. **M.M. Nickel:** None. **T.R. Tölle:** None. **M. Ploner:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.02/FF16

Topic: D.03. Somatosensation: Pain

Support: DFG PL 321/10-1

DFG PL 321/11-1

Title: Prefrontal gamma oscillations encode spontaneous fluctuations of chronic back pain intensity

Authors: *E. S. MAY[1], M. M. NICKEL[1], S. TA DINH[1], L. TIEMANN[1], H. HEITMANN[1], I. VOTH[2], T. R. TÖLLE[2], J. GROSS[3], M. PLONER[1]
[1]Dept. of Neurol. and TUM-Neuroimaging Ctr., [2]Dept. of Neurol., Technische Univ. München, Muenchen, Germany; [3]Inst. of Neurosci. and Psychology, Univ. of Glasgow, Glasgow, United Kingdom

Abstract: Chronic pain is a major health care issue, whose neural basis is still incompletely understood. Previous work using functional magnetic resonance imaging has implicated the medial prefrontal cortex in chronic back pain, which has been associated with negative emotions and the affective value of sensory stimuli. In addition, studies using electroencephalography (EEG) revealed a role of prefrontal gamma oscillations in the encoding of the subjective intensity of tonic experimental pain in healthy subjects. However, the role of such higher frequency oscillatory brain activity in chronic pain has not yet been studied. In the current study, we asked 27 chronic back pain patients to continuously monitor and rate their current pain intensity while simultaneously recording EEG. In line with previous studies, behavioral data showed fluctuations of pain intensity within 10 minutes of recording. Time-frequency analyses of brain activity and cluster-based permutation statistics showed a positive relation between current pain intensity and the amplitude of prefrontal gamma oscillations (30-100 Hz, $p = 0.002$), but no associations between pain intensity and brain activity in theta (4-7 Hz), alpha (8-13 Hz) or beta (14-29 Hz) frequency bands. This relation was not present in a visual control condition controlling for sensory, motor and attentional components of the continuous pain rating procedure. These results indicate that chronic back pain intensity is encoded by prefrontal

gamma oscillations, revealing a potential frequency and spatially specific functional brain marker of chronic pain intensity. This finding extends previous work in healthy subjects, which showed that prefrontal gamma oscillations encode the subjective intensity of tonic experimental pain, while the objective stimulus intensity is reflected in sensorimotor alpha and beta oscillations. Thus, the current results are in line with a detachment of the neural processes underlying chronic pain from early sensory processes and a shift towards emotional-evaluative neural circuits.

Disclosures: **E.S. May:** None. **M.M. Nickel:** None. **S. Ta Dinh:** None. **L. Tiemann:** None. **H. Heitmann:** None. **I. Voth:** None. **T.R. Tölle:** None. **J. Gross:** None. **M. Ploner:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.03/FF17

Topic: D.03. Somatosensation: Pain

Support: DFG PL 321/10-1

DFG PL 321/10-2

DFG PL 321/11-1

Title: Brain networks of chronic pain assessed by resting-state EEG

Authors: *S. TA DINH[1], L. TIEMANN[1], M. M. NICKEL[1], E. S. MAY[1], H. HEITMANN[1], D. UTPADEL-FISCHLER[2], G. EDENHARTER[2], T. R. TÖLLE[2], J. GROSS[3], M. PLONER[1]
[1]Dept. of Neurol. and TUM-Neuroimaging Ctr., [2]Dept. of Neurol., Technische Univ. München, Muenchen, Germany; [3]Inst. of Neurosci. and Psychology, Univ. of Glasgow, Glasgow, United Kingdom

Abstract: Chronic pain is a major healthcare issue whose causes are not fully understood and whose treatment is often unsatisfactory. Recent evidence has shown that the brain plays an important role in the development and maintenance of chronic pain. Magnetic resonance imaging has shown structural and functional changes in the brains of chronic pain patients at rest in different brain networks at frequencies below 0.1 Hz. Whether changes of brain networks also occur at higher frequencies assessed by electrophysiological recordings has remained largely unknown. We have recorded electroencephalography (EEG) during restful wakefulness in 20 chronic pain patients suffering from Fibromyalgia and 22 matched healthy controls. In accordance with previous findings, we observed a trend towards increased frontal brain activity at theta (4 - 8 Hz) frequencies in chronic pain patients. To assess potential changes of brain activity at the network level, we have performed graph theory-based network analyses of EEG

data at theta (4 - 8 Hz), alpha (8 - 13 Hz), beta (14 - 30 Hz) and gamma (60 - 100 Hz) frequencies. Specifically, we investigated the following graph measures: degree centrality, clustering coefficient, global efficiency, small-worldness, modularity and the global hub disruption index. Network analyses based on functional connectivity measured by the debiased weighted phase lag index were evaluated using permutation statistics and did not differ significantly between groups, neither at electrode nor source level. These results will be complemented by an extension of the analyses to a larger cohort of 100 chronic pain patients with respective controls and the amplitude envelope correlation as an additional measure of functional connectivity. As chronic pain affects many domains of sensory, cognitive and affective functioning we expect to find changes on a global network scale. These analyses promise to further the understanding of the brain mechanisms of chronic pain and could help to define brain- based biological markers of pain assessed by portable and affordable devices.

Disclosures: S. Ta Dinh: None. **L. Tiemann:** None. **M.M. Nickel:** None. **E.S. May:** None. **H. Heitmann:** None. **D. Utpadel-Fischler:** None. **G. Edenharter:** None. **T.R. Tölle:** None. **J. Gross:** None. **M. Ploner:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.04/FF18

Topic: D.03. Somatosensation: Pain

Support: NSERC Discovery grant

CIHR Canada Research Chair Program

Canadian Fund for Innovation (CFI)

Dalhousie University

Title: Pain modulation; when expectations become the perception to maintain the status quo

Authors: M. LIM[1], A. MCINTYRE[2], C. O'GRADY[3], M. LYNCH[4], S. MATWIN[5], S. BEYEA[5], *J. A. HASHMI[4]
[1]Anesthesia, Pain Mgmt. and Perioperative Med., [2]Fac. of Computer Sci., [3]Physics and Atmospheric Sci., [4]Anesthesia, Pain Mangement and Perioperative Med., [5]Dalhousie Univ., Halifax, NS, Canada

Abstract: It is known that expectations induced with top-down visual cues can alter perceived pain intensity. Expectations are mental representations of internal models of our external realities. The internal model guides how new sensory inputs will be organized and interpreted

during the perceptual process. However, it is unclear how sensory decisions and brain activity map deviations in the associative rule that links top-down cues with bottom-up stimuli.
We hypothesized that learning a linear associative rule between cue and stimulus is preserved even when bottom-up stimulus intensity is altered and when top-down information in cues is missing.
16 healthy participants (8 women) were trained with probabilistic cues (0-100) that were linearly related with 10 incrementing heat stimulus intensities in a randomized order. The linear association between cue and stimulus was tested by inverting cue-stimulus association in events representing three conditions (A-C). A. values from a low cue range (0-40) and a high cue range (60-100) were coupled with a single low (45°) or high (47°) stimulus intensity respectively. B. values from a low cue range were coupled with a high stimulus intensity and C. top-down information in cue was absent and bottom-up stimuli were held at high or low intensity.
As hypothesized, pain ratings showed a linear relation with cues even when the stimulus intensity was held at two levels (condition A; $P < 0.007$). In condition B. pain was evaluated as significantly less painful ($P < 0.001$) when the high stimulus was coupled with cues selected from a low range versus a high range. In condition C. the evaluation of low and high stimuli in the absence of top-down cues was not significantly different ($P = 0.062$) from evaluations of these stimuli observed during the training phase. Neural responses mirrored the pattern of pain responses in conditions A and B. showing a linear relationship between cues and brain activations ($p<0.05$ corrected) with peak effects in dorsal ACC and right anterior insula.
Our findings demonstrate that pain is modulated by expectations that result from an internal model established through learning associative rules. Thus, even when bottom-up sensory inputs are significantly modified, the perceived pain is modulated to fit with the rules established in the internal model. The results offer new insights on the subjective nature of pain and show a bias in pain perception towards top-down information and past experiences.

Disclosures: **M. Lim:** None. **A. McIntyre:** None. **C. O'Grady:** None. **M. Lynch:** None. **S. Matwin:** None. **S. Beyea:** None. **J.A. Hashmi:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.05/FF19

Topic: D.03. Somatosensation: Pain

Support: DFG PL 321/10-1

DFG PL 321/10-2

DFG PL 321/11-1

Title: Brain networks of tonic experimental pain assessed by EEG

Authors: *M. M. NICKEL, S. TA DINH, E. S. MAY, L. TIEMANN, M. POSTORINO, M. PLONER
Dept. of Neurol. and TUM-Neuroimaging Ctr., Technische Univ. München, Muenchen, Germany

Abstract: Pain is a complex phenomenon which results from the integration of sensory and a broad variety of contextual processes. In the brain, the complexity of pain is reflected by activations of an extended network of brain areas displaying oscillations and synchrony at different frequencies. However, brain networks of pain have mostly been investigated in relation to brief experimental pain stimuli whereas the main clinical problem of chronic pain is characterized by longer-lasting pain. We have therefore recorded electroencephalography (EEG) during 10 minutes of tonic heat pain applied to the left and right hand in 39 healthy volunteers. To assess tonic pain-related brain activity at the network level, we performed graph theory-based network analyses of EEG data at theta (4-8 Hz), alpha (8-13 Hz), beta (14-30 Hz) and gamma (60-100 Hz) frequencies. Networks were constructed based on functional connectivity measured by the debiased weighted phase lag index on electrode level. The network measures degree centrality, clustering coefficient, global efficiency and small-worldness were compared between the tonic pain and a control condition using permutation statistics. We observed trends towards decreased small-worldness at alpha and gamma frequencies and a decreased global clustering coefficient at gamma frequencies during tonic pain. Additionally, in tonic pain conditions, a trend towards a local decrease in degree centrality was found at fronto-central electrodes at theta frequencies, which was consistent across the two hands. Thus, first analyses hinted at both global and local alterations of the functional network structure during the perception of tonic pain. These results will be extended by source level analyses and the application of complementary measures of functional connectivity, e.g. the amplitude envelope correlation. The analyses will broaden the understanding of the brain mechanisms of pain to the network representation of longer-lasting pain as a hallmark of chronic pain syndromes.

Disclosures: M.M. Nickel: None. **S. Ta Dinh:** None. **E.S. May:** None. **L. Tiemann:** None. **M. Postorino:** None. **M. Ploner:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.06/FF20

Topic: D.03. Somatosensation: Pain

Support: DFG PL 321/11-1

DFG PL 321/13-1

Title: Neurophysiological mediators of the perceptual, autonomic and behavioral components of pain

Authors: L. TIEMANN, V. D. HOHN, E. S. MAY, M. M. NICKEL, S. TA DINH, *M. PLONER
Neurol., TU Muenchen, Munich, Germany

Abstract: Pain is a complex phenomenon which translates sensory information into perception and autonomic and behavioral responses in order to protect the physical integrity of the individual. In the brain, the complexity of pain is reflected by patterns of pain-associated neural activity in different brain areas and at different latencies and frequencies. However, whether and how these different brain responses differentially contribute to the perceptual, autonomic and behavioral components of pain has remained largely unknown yet. In the current electroencephalography (EEG) experiment, we addressed this question by examining the neurophysiological responses evoked by noxious cutaneous laser stimuli of different intensities and relating them to perceptual (pain rating), behavioral (reaction time) and autonomic (skin conductance) pain responses recorded in 51 healthy subjects. Data were analyzed by means of multi-level mediation analysis which provides a unique opportunity to quantify the degree to which the relationship between objective stimulus intensity (independent variable) and subjective rating, reaction times or skin conductance (dependent variables) can be explained by different types of brain activity (mediators). The results show that the N1, N2 and P2 components of the well-known pain-evoked potential as well as activity in the theta frequency band (4-7 Hz) significantly mediated the effect of stimulus intensity on both pain ratings and reaction times. In contrast, responses at gamma frequencies (70-90 Hz) significantly mediated the effect of stimulus intensity on reaction times only, indicating a dissociation between the neurophysiological processes underlying the perceptual and behavioral components of pain. The neural mediators of skin conductance responses are currently being investigated. These results show how the present approach can comprehensively characterize the neurophysiological processes underlying the translation of noxious stimuli into the different components of pain. Beyond, the experiment showcases a novel approach to further the understanding of the functional significance of pain-associated EEG responses in health and disease.

Disclosures: L. Tiemann: None. **V.D. Hohn:** None. **E.S. May:** None. **M.M. Nickel:** None. **S. Ta Dinh:** None. **M. Ploner:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.07/FF21

Topic: D.03. Somatosensation: Pain

Support: NIH SCCIH (P01-AT006663)

Korean Institute of Oriental Medicine

Title: Functional connectivity between the somatosensory representation of the back and Salience Network areas encode clinical pain in chronic low back pain patients

Authors: J. KIM[1], I. MAWLA[2], J. KONG[2], J. LEE[2], J. GERBER[2], C. JUNG[2], A. ORTIZ[2], S.-T. CHAN[2], *M. L. LOGGIA[2], A. D. WASAN[3], R. EDWARDS[4], R. L. GOLLUB[2], B. ROSEN[2], V. NAPADOW[2]
[1]Korea Advanced Inst. of Sci. and Technol., Daejeon, Korea, Republic of; [2]Radiology, Massachusetts Gen. Hosp. / Harvard Med. Sch., Charlestown, MA; [3]Dept. of Anesthesiol., Ctr. for Pain Research, Univ. of Pittsburgh, Pittsburgh, PA; [4]Dept. of Anesthesiology, Perioperative and Pain Med., Brigham and Women's Hosp., Boston, MA

Abstract: Despite evidence for structural and functional brain changes in chronic low back pain (cLBP) patients, the relation of these changes to the perception of clinical pain is still not clear. We previously investigated the role of primary somatosensory cortex (S1) functional connectivity (Kim et al. A&R 2015) in the brain processing of clinical pain and hyperalgesia in patients with chronic pain. In this study, we investigated functional connectivity between S1 and brain regions within the Salience Network, and how it is related to clinical pain processing within a large cohort of cLBP patients. BOLD fMRI data from 133 cLBP patients (age=40.3±11.8 yo, 73 F) and 54 healthy controls (HC, age=40.4±11.0 yo, 30 F) were used for resting connectivity analyses (Siemens 3T, TR=3s, 120 volumes). For cLBP, resting fMRI data were collected before and after individually-tailored low back pain exacerbation maneuvers (e.g. sit-ups, toe-touching, and twisting). Additionally, event-related fMRI runs with intermittent painful electrical stimulation applied to low back were used to localize the S1 subregion of the low back for seed definition. Clinical back pain in cLBP was significantly increased after pain exacerbation (34.0±21.9 vs. 55.2±23.0, $p<0.001$). Compared to HC, patients exhibited increased connectivity between salience network and a S1 cluster overlapping the low back representation (as determined by the experimental electrical pain fMRI data). Connectivity was increased relative to HC for cLBP both before and after clinical pain exacerbation. In turn, S1-seed based connectivity was increased to insula cortex and thalamus, compared to HC. Within the cLBP, clinical maneuvers increased S1 connectivity to the temporoparietal junction, dorsolateral prefrontal cortex and bilateral thalamus. Furthermore, a whole-brain linear regression analysis found that increased S1 connectivity to anterior insula, thalamus and midbrain was correlated with increasing clinical pain, closely linking back representation S1-specific connectivity with clinical pain perception. Our results of cLBP patients suggest that increased functional connectivity between the S1 representation of the low back and salience processing regions such as anterior insula as well as thalamus encodes clinical pain perception. Determination of the neural correlates of clinical pain has been elusive so far, and functional connectivity between body region-specific somatosensory and salience processing brain areas may serve as a key neurophysiological substrate for emergent clinical pain in chronic pain patients.

Disclosures: **J. Kim:** None. **I. Mawla:** None. **J. Kong:** None. **J. Lee:** None. **J. Gerber:** None. **C. Jung:** None. **A. Ortiz:** None. **S. Chan:** None. **M.L. Loggia:** None. **A.D. Wasan:** None. **R. Edwards:** None. **R.L. Gollub:** None. **B. Rosen:** None. **V. Napadow:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.08/FF22

Topic: D.03. Somatosensation: Pain

Support: NIH Grant P30 NR014129

Title: Brush allodynia and mechanical hyperalgesia: Predictors and associations

Authors: Y. LIU[1,2], M. L. KEASER[1], A.-C. SCHMID[3], T. J. MEEKER[1], S. G. DORSEY[4], D. A. SEMINOWICZ[1], *J. D. GREENSPAN[1]
[1]Dept Neural and Pain Sci., Univ. Maryland Dent. Sch., Baltimore, MD; [2]Dept Pain Med., Peking Univ., Beijing, China; [3]Dept Clin. Neuroengineering, Swiss Federal Inst. of Technol. (EPFL), Geneva, Switzerland; [4]Dept Pain and Translational Symptom Sci., Univ. Maryland Sch. of Nursing, Baltimore, MD

Abstract: Introduction: Brush allodynia (BA) often, but variably occurs in many chronic neuropathic pain conditions. What leads to the development of BA in any given individual is unknown. Secondarily, it is not clear how strongly BA is related to mechanical hyperalgesia. The goal of this study was to determine: 1) what factors are associated with capsaicin-induced BA and mechanical hyperalgesia susceptibility, and 2) the extent to which a sensitizing provocation separately engages hyperalgesia and allodynia. **Methods**: Fifty-four healthy participants (24F) between 18-43 years (27.7±6.0) were evaluated before and after exposure to a sensitizing capsaicin-heat pain model (C-HP). The model consisted of topical application of 10% capsaicin cream, accompanied by a heat stimulus for 35 minutes provoking a moderate level of pain. Sensory testing included warmth detection thresholds (WDT), heat pain thresholds (HPT), and Mechanical Pain Ratings (MPR) of sharp probes of various intensities. Additionally, the following questionnaires were administered: Pain Sensitivity Questionnaire (PSQ), Beck Depression Inventory (BDI); State-trait anxiety (STAI), and the PILL to measure somatic awareness. **Results**: Predictors of brush allodynia: Lower WDT and HPT; higher ratings of punctate pain at low levels of stimulation (near threshold). Predictors of mechanical hyperalgesia: 1) *Hyperalgesia to lower intensity stimuli*: Lower HPT; higher ratings to all levels of punctate probe stimulation; higher scores on PSQ. 2) *Hyperalgesia to higher intensity stimuli*: higher scores on psychological measures of depression, anxiety, and somatic awareness. Associations between allodynia and mechanical hyperalgesia: Those with allodynia showed greater hyperalgesia than those without allodynia, most strongly at the first post-exposure time

point. In contrast, the area of allodynia did not correlate with the area of hyperalgesia at any time point. **Conclusions**: Greater sensitivity to heat and punctate mechanical stimuli near pain threshold significantly separated the BA susceptible from those not developing BA with the C-HP model. Greater baseline sensory sensitivity to painful stimuli predicted greater mechanical hyperalgesia for lower intensity stimuli, but not for higher intensity stimuli. Instead, psychological factors, rather than sensory sensitivity, were predictive of mechanical hyperalgesia at stronger levels of stimulation. These data suggest that factors driving sensitization-induced mechanical hyperalgesia differ at lower vs. higher stimulation levels, and only the former have overlap with allodynic factors.

Disclosures: Y. Liu: None. **M.L. Keaser:** None. **A. Schmid:** None. **T.J. Meeker:** None. **S.G. Dorsey:** None. **D.A. Seminowicz:** None. **J.D. Greenspan:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.09/GG1

Topic: D.03. Somatosensation: Pain

Support: UAB Neuroimaging Program Development and Research Acceleration Awards 2015

Title: Functional diffusion tensor imaging (fDTI) of the human spinal cord during painful thermal stimulation

Authors: *S. C. MUELLER, J. C. LIN, J. W. YOUNGER
Psychology, Univ. of Alabama At Birmingham, Birmingham, AL

Abstract: Introduction: The experience of pain involves both ascending nociceptive signals and descending inhibitory controls, both travelling through the spinal cord. These processes are difficult to measure in living humans, in part because non-invasive methods for imaging long axon activity are lacking. We are developing a modification of diffusion tensor imaging (DTI) for the measurement of task-related changes in axon activity via slow changes in spinal cord diffusion (fractional anisotropy; FA). We are first applying this technique to measuring spinal cord activity changes to pain stimuli. The tract of interest was the afferent spino-thalamic tract (STT), which is involved in relaying peripheral pain signals to the brain. We expected increases in mean FA after onset of the pain stimulus in the STT contralateral to stimulation, indicating increased spinal transmission of nociceptive messages.
Materials & Methods: We acquired structural (T2-weighted) and functional (diffusion weighted) images of spinal cords in 19 healthy males during a pain stimulation task. Participants received 4 blocks of the following: 5 minutes of baseline, 1 minute of thermal stimulus to the thenar eminence of the left hand (dermatome C6), and 4 minutes of recovery. Diffusion data was

registered to the T2-weighted image, and FA values from tracts of interest extracted. Generalized Estimating Equations (GEEs) were used to contrast baseline activity to 3 minutes post-pain-onset across the 4 blocks. GEEs were constructed for each tract of interest (STT ipsilateral and contralateral to the noxious stimulus). Activity was also tested above and below the C6 dermatome insertion point.
Results: GEEs indicated significant task-related decreases in FA in the contralateral STT. The task-related change required approximately 1 minute to peak, and 2 minutes to return to baseline. Changes were observed at spinal levels above the peripheral insertion point (C6), while activity below C6 remained stable.
Conclusion: FA values indicate the degree to which water diffusion in the spinal cord is restricted to one direction. A decrease suggests less restricted diffusion in the contralateral STT during painful stimulation, which may be due to morphological changes of glial cells. Notably, the task-related activity operated in a timescale (3 minutes) much different than that of blood-oxygen dependent (BOLD) measures (a few seconds). With further development, this technique may allow researchers to investigate normal and pathological pain transmission in the spinal cord.

Disclosures: S.C. Mueller: None. **J.C. Lin:** None. **J.W. Younger:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.10/GG2

Topic: D.03. Somatosensation: Pain

Title: How'd you do? The impact of openness during competition on perceived pain in women

Authors: *A. B. SIMMONS, C. DE GUZMAN, J. J. SOLIS, J. R. VILLATORO, J. BOYETTE-DAVIS
Behavioral Neurosci., St. Edward's Univ., Austin, TX

Abstract: There are various influences on pain perception in women, including personality traits. Previous research has found that neuroticism and openness to experience are positively correlated with pain, but this did not account for levels of sex hormones, an important factor in pain perception in women. The current study investigated the impact of personality and testosterone levels in women experiencing a painful stimulus. Pain was induced in participants (n=22 women) using the cold pressor test (CPT) wherein participants placed their hand in water that was maintained at 2^0 C and rated the amount of pain perceived using a visual analog scale (VAS). Tolerance was captured as the amount of time participants kept their hand in the water. To further examine the influence of testosterone, half of the participants were led to believe that others exposed to the CPT were able to withstand the CPT-induced pain for the duration of the

test session; the other half were not provided with any such information, thereby creating competition and no-competition groups. The results indicated that when competition was present, participant VAS was significantly negatively correlated to the amount of time spent in the CPT (r= -.64, p= .03). Further, those scoring higher in openness spent more time in the CPT (r=.69, p=.04). These results were not found in the no-competition group. These data indicate that higher levels of openness when in a competitive environment increase pain tolerance, possibly in relation to changes in testosterone, clarifying previous research investigating personality traits in female pain perception.

Disclosures: A.B. Simmons: None. **C. de Guzman:** None. **J.J. Solis:** None. **J.R. Villatoro:** None. **J. Boyette-Davis:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.11/GG3

Topic: D.03. Somatosensation: Pain

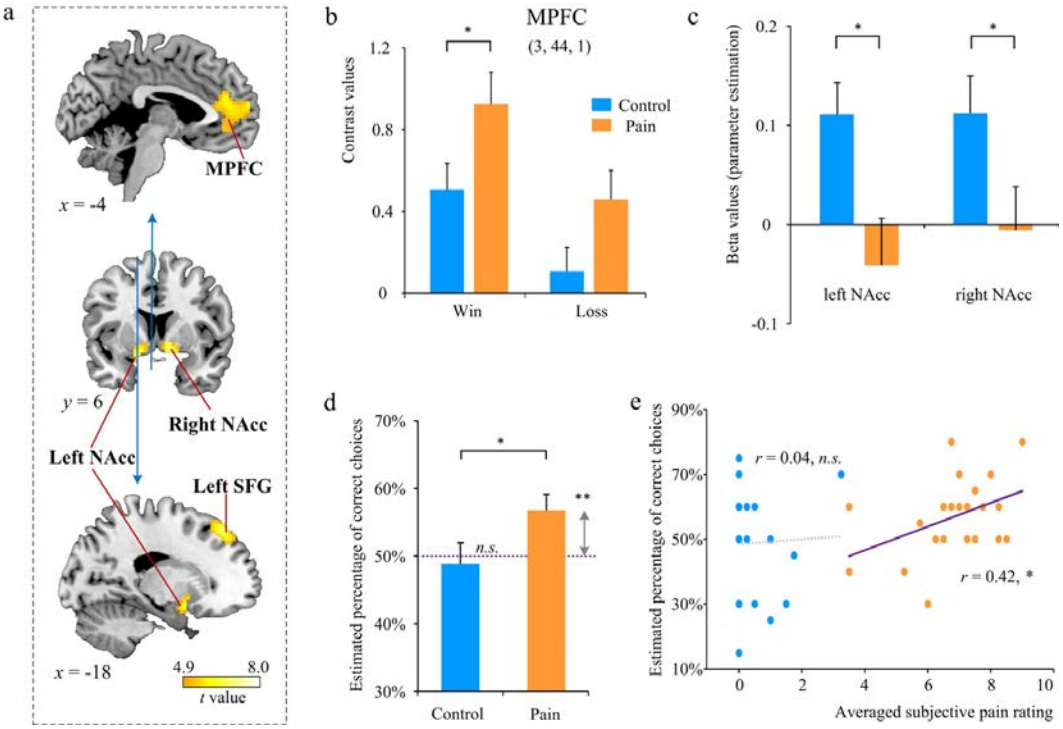
Support: National Natural Science Foundation of China (31600890)

Title: Physical pain enhances reward-related brain activation in medial prefrontal cortex

Authors: *C. WANG, J. GAO, X.-W. DONG
East China Normal Univ., Shanghai City, China

Abstract: Pain modulates motivation and motivation-related brain activities. For example, individuals in physical pain are motivated to escape from aversive states. However, it is unclear about the mechanism of how pain modulates motivation in humans. Animal studies indicate that the disturbed reward neural circuitry by chronic pain may responsible for motivation change. By using fMRI technology, the present study aimed to investigate the modulation of physical pain on brain activities in the reward neural circuitry. A total of 60 healthy university students were recruited and then were treated with either Capzasin (pain group) or hand cream (control group). During brain imaging, they played a card guess game in which they would receive monetary gain by a correct guess or monetary loss by an incorrect guess. Results showed that, the medial prefrontal cortex (MPFC) and bilateral nucleus accumbens (NAcc) were activated when individuals won during the card guess game in both groups (Fig. a). Interestingly, individuals in the physical pain group relative to those in the control group showed greater activation in the MPFC in the win condition (Fig. b), indicating that physical pain may increase the expectation of monetary reward. The MPFC-NAcc connectivity was reduced by physical pain (Fig. c). Moreover, individuals in pain group possessed a positive estimation of their performance (Fig. d) and such positive bias was correlated with subjective rating of pain intensity (Fig. e). Our

findings suggest that physical acute pain may increase reward-related MPFC activation and such facilitation of brain activity might be associated with a high expectation of reward and a positive bias of performance estimation. The study provides a potential mechanism of how pain modulates motivation, and sheds light on the consequences of pain on human cognition and behavior.



Disclosures: **C. Wang:** None. **J. Gao:** None. **X. Dong:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.12/GG4

Topic: D.03. Somatosensation: Pain

Support: NIH/NINDS R01-NS035115-12

NIH/NINDS R01-DE022746-01

Title: Hippocampal subfield volumes remain time invariant in chronic pain and transition to chronic pain

Authors: *T. B. ABDULLAH, A. T. BARIA, L. HUANG, A. V. APKARIAN
Physiol., Northwestern Univ. Feinberg Sch. of Medicin, Chicago, IL

Abstract: The hippocampal formation is a complex, heterogeneous, structure which can be segregated into functional units along its longitudinal axis. The growing consensus asserts that the dorsal/posterio r component is associated with cognitive processes, while the ventral/anterior hippocampus is associated with emotional and stress related functions. Here we aimed to determine how the volume and functional connectivity of hippocampus subcomponents along the anteroposterior axis, as well as, along the neuroanatomical divisions of the hippocampal formation relate to different kinds of clinical pain. We collected T1 images and resting-state fMRI from 23 healthy controls, 22 chronic low back pain patients, 37 sub-acute persisting and 27 sub-acute recovering low back pain patients across 4 time points over a one-year period. We extracted whole hippocampal and subfield volumes using automated segmentation tools in FreeSurfer. All structural volumes were corrected for whole brain volume excluding ventricles. Functional images were motion corrected with ICA-AROMA and bandpass-filtered at 0.01 - 0.08 Hz. Functional connectivity was performed only in healthy controls, using a seed-based analysis with four regions of interests (ROIs) in bilateral anterior and poster hippocampi (antHC and postHC). Seeds were uniquely determined for each subject by FreeSurfer anatomical parcellation, and average time series from these regions were fit to all voxels in the brain (excluding hippocampal voxels) using a general linear model to generate functional connectivity maps. A two-way repeated measure ANOVA on subfield volume for time and pain-type yielded a significant time effect only in the right CA1-CA3 area ($F_{(3,315)} = 3.84$, $p < 0.01$), showing a general decrease in volume over one year which was not specific to any pain type. There was no significant condition-time interaction. Connectivity differences between ant/post HC indicated stronger antHC connectivity to ipsilateral parahippocamapl gyri, and stronger postHC to posterior cingulate cortex, precuneus, and visual cortices. Overall, our results indicate changes in hippocampal volumes are not implicated in transition or maintenance of chronic pain. Additionally, there is distinct functional differentiation of the antHC and postHC. How this differential hippocampal connectivity manifests in pain will be addressed. Funded by: NIH/NINDS R01-NS035115-12 and R01-DE022746-01

Disclosures: T.B. Abdullah: None. **A.T. Baria:** None. **L. Huang:** None. **A.V. Apkarian:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.13/GG5

Topic: D.03. Somatosensation: Pain

Support: Natural Science and Engineering research council (NSERC) of Canada

FRQS

CFI

Title: Integration of bilateral nociceptive signals: When more is less

Authors: *S. NORTHON, *S. NORTHON, N. RUSTAMOV, S. BOIS, S. SCHREIBER, M. PICHE
Dept. de Chiropratique, Univ. Du Quebec A Trois-Rivieres, Trois-Rivieres, QC, Canada

Abstract: Integration of multiple concurrent somatosensory signals is an essential process to promote adapted behaviors in response to multiple stimuli from the environment. For instance, tactile inputs on fingers of both hands are integrated at early stages of sensory processing (response suppression, as measured with magnetoencephalography), which may promote coordinated bimanual tasks. To gain insight into cortical processing and integration of concurrent nociceptive signals, laser heat stimuli can be used to recruit nociceptors selectively. The most common measure of laser-evoked potentials (LEPs), namely the N2-P2, reflects activity in multiple brain regions involved in the central processing of nociceptive inputs. The aim of the present study was to examine the integration of bilateral nociceptive signals. Nineteen healthy right-handed participants (9 F, age: 25 ± 4) were recruited. Nociceptive stimuli were produced by a Nd :YAP laser system. Cortical activity was recorded with a 64-channel EEG system. Four counterbalanced blocks (20 trials each, 6 s inter-stimulus interval) were performed, during which stimuli were applied to the right or left hand or to both hands concurrently with attention to the right or left. The N2-P2 was measured at the vertex (Cz) and its amplitude was compared between the 4 blocks. N2P2 amplitude was not significantly different between the unilateral right vs left condition ($p=0.6$) and between the bilateral conditions with attention to the right or left ($p=0.22$). Also, the N2-P2 amplitude was decreased in bilateral conditions (attention to right or left) compared with their respective unilateral condition, (right: $p<0.001$; left: $p=0.013$). These results are consistent with a previous study on tactile integration and indicate that integration of concurrent bilateral nociceptive signals in the central nervous system is reflected by reduced N2P2 amplitude. This effect was not affected by spatial attention and may reflect a relative decrease of saliency for each stimulus.

Disclosures: S. Northon: None. **N. Rustamov:** None. **S. Bois:** None. **S. Schreiber:** None. **M. Piche:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.14/GG6

Topic: D.03. Somatosensation: Pain

Support: 'Strategic Priority Research Program (B)', CAS, Grant XDB02010400

Title: Pain sensation and thermal pain-evoked fMRI activity changes in chronic low back pain patients

Authors: *Q. YANG[1], L.-X. YANG[1,2], Y.-H. XU[1,2], L. CHEN[1,3,4]
[1]Shanghai Clin. Res. Ctr., Shanghai Inst. for Biol. Sciences, CAS, Shanghai, China; [2]Xuhui Central Hosp., Shanghai, China; [3]Dept. of Radiology and Radiological Sci., [4]Inst. of Imaging Sci., Vanderbilt Univ., Nashville, TN

Abstract: This study aims to characterize psychophysical measures of heat pain changes in chronic low back pain patients (CLBP); and to investigate CLBP's brain activities responded to thermal pain.

Methods

We recruited 101 CLBP and 100 healthy controls (CON) with their written informed consents. Each participant went through a psychophysical test prior to MRI scans. Participants reported their pain levels (numeric scale 0-10) from mild, moderate, to severe while ramping temperatures (baseline=32°C; ascending rate=0.5°C/s; 3 repeats) were delivered to their lower back skin with an ATS thermode (Medoc PATHWAY). Participants were scanned in a 3-T Siemens MRI system with a 32-channel head coil. T2*-weighted fMRI were acquired (EPI; TR = 2 s, voxel size = 3×3×5 mm^3, duration = 10 mins). During this scan, thermal stimuli (Fig. A; temperature = 43, 46 and 47°C; durations 8-23 s) were applied to the back of participants. Participants rated their pain with a finger-span device and a visual feedback.

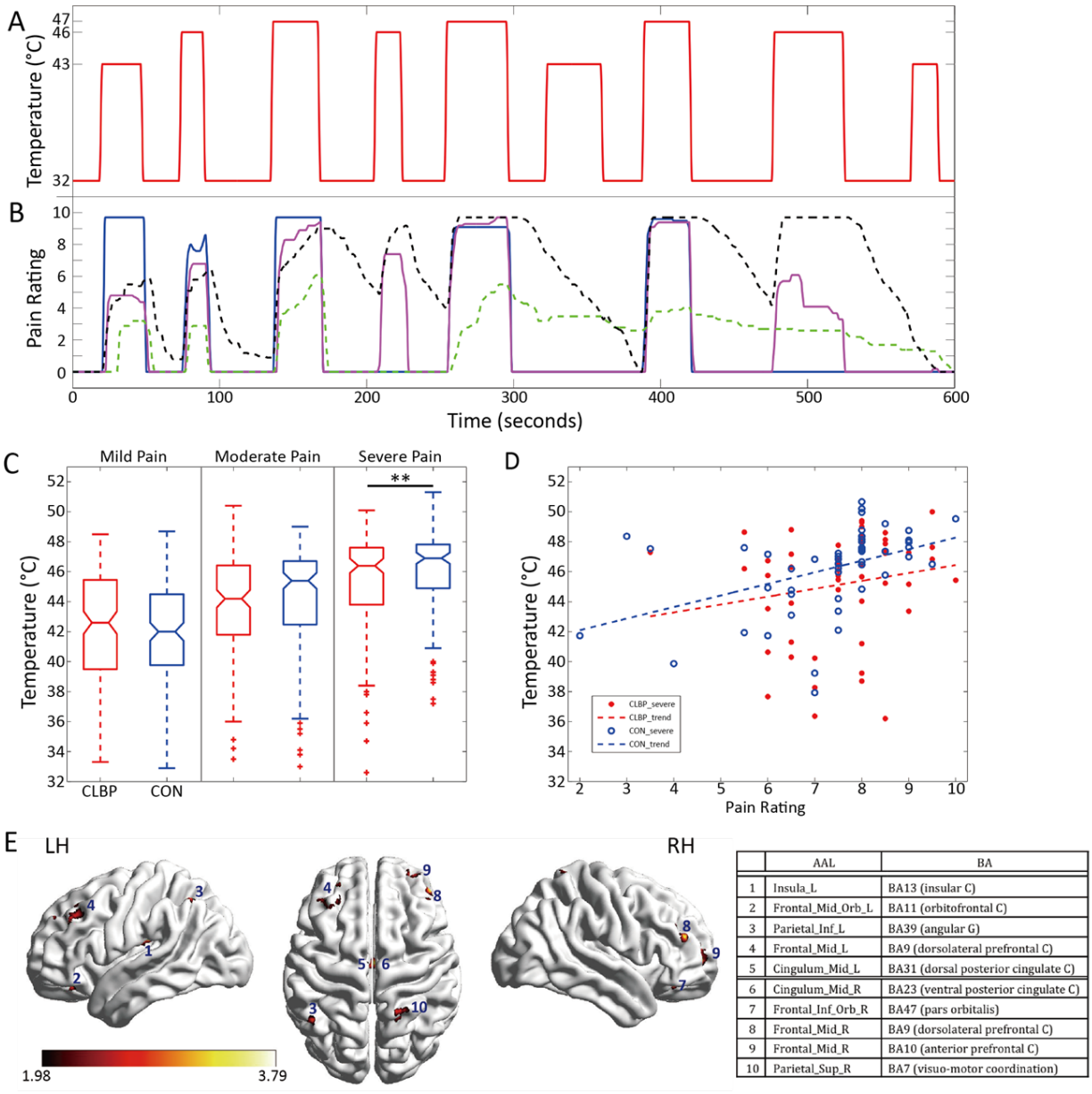
Participants owned more than three white matter lesions (diagnosed by two radiologists), or had inadequate real-time pain rating (correlation < 0.55; Fig. B) were excluded. Finally, 53 pairs of CLBP (age = 53.4 ± 9.8 years; mean pain history = 5.3 years) and age-/gender-matched CON were included. Statistics of psychophysical tests were examined in MATLAB. Thermal stimulus-evoked fMRI changes between CLBP and CON were compared (SPM 8).

Results

Psychophysical analysis showed that CLBP reported significantly lower temperature for severe pain ($p < 0.01$, Fig. C) than CON. Additionally, the examination of temperature-rating for severe pain revealed that CLBP tended to rate higher for the lower temperature (dotted lines, Fig. D). Compared to CON, 10 brain areas in the CLBP exhibited significantly higher fMRI signals (Fig. E), including e.g. left insular (1), left orbitofrontal (2) bilateral cingulate (5, 6) and dorsolateral prefrontal (4, 8) cortex.

Conclusions

CLBP showed altered pain sensitivities for severe heat pain. CLBP had widespread changes in fMRI detected in pain-, memory- and cognition-related regions.



	AAL	BA
1	Insula_L	BA13 (insular C)
2	Frontal_Mid_Orb_L	BA11 (orbitofrontal C)
3	Parietal_Inf_L	BA39 (angular G)
4	Frontal_Mid_L	BA9 (dorsolateral prefrontal C)
5	Cingulum_Mid_L	BA31 (dorsal posterior cingulate C)
6	Cingulum_Mid_R	BA23 (ventral posterior cingulate C)
7	Frontal_Inf_Orb_R	BA47 (pars orbitalis)
8	Frontal_Mid_R	BA9 (dorsolateral prefrontal C)
9	Frontal_Mid_R	BA10 (anterior prefrontal C)
10	Parietal_Sup_R	BA7 (visuo-motor coordination)

Figure: (A) Temperature profile of thermal stimuli applied to the back of participants during their fMRI scans. (B) Examples of real-time pain rating of 2 participants. The blue and magenta lines well synchronized with (A); black and green dotted lines poorly correlated with (A), thus these participants were excluded from further analyses. (C) Temperatures recorded for three-level hot pain stimuli (mild, moderate and severe) of CLBP (red) and CON (blue) groups (53 pairs; **: $p < 0.01$). (D) Temperatures reported for severe pain with the scores given by each individual (red: CLBP; blue: CON). (E) Ten brain areas showed significantly different activations for heat pain between CLBP and CON (53 pairs; two-sample double-tailed t-test, thresholded at $p < 0.05$, peak differences $p < 0.01$ and cluster areas > 27 voxels).

Disclosures: **Q. Yang:** None. **L. Yang:** None. **Y. Xu:** None. **L. Chen:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.15/GG7

Topic: D.03. Somatosensation: Pain

Support: Seed Grant BYU MRIRF

Title: An fMRI analysis of peripheral neuropathy pain before and after treatment with transcutaneous electrical nerve stimulation

Authors: *T. BODILY[1], J. PEACOCK[1], D. BUSATH[2], B. KIRWAN[3]
[2]PdBio, [3]Psychology, [1]Brigham Young Univ., Provo, UT

Abstract: Chronic pain is a widespread and costly pathology. Treatment regimens are typically only marginally successful in the long-term, especially when pain etiology is neuropathic. Transcutaneous electrical nerve stimulation (TENS) has been shown to be effective as an analgesic for some acute and musculoskeletal chronic pain conditions, but evidence regarding TENS efficacy in treating chronic neuropathic pain is unclear. The neural signature of chronic pain, thought to be localized largely in emotional and affective brain regions, is not well characterized for many chronic pain conditions, including peripheral neuropathy. Accordingly, we performed a treatment trial of two TENS protocols in subjects with peripheral neuropathies with pre- and post-treatment functional MRI scans.

Eighteen subjects were recruited from a pool of interested candidates by screening for compatible medication history, a minimum average pain level of 4/10, and a report of peripheral neuropathy symptoms. Subjects were randomly assigned to either of the two TENS treatment protocols and were given one thirty-minute treatment session. A double blind of therapist and subject awareness of treatment protocol was maintained throughout the duration of the study. Participants underwent three MRI scanning sessions--one immediately before the treatment, one immediately after, and one on the following day. Scan sessions included both structural and resting state fMRI. After removal of the blind, participants from one of the TENS protocols were invited to return for a crossover treatment with an MRI scan sequence parallel to the first treatment.

Both groups reported lower pain immediately after treatment, and pain levels in both groups returned to baseline by the following day. The crossover subjects did not experience a greater pain reduction from either treatment. Graph theoretical analysis of the resting state fMRI data revealed a decrease in global network efficiency across all subjects from pre- to post-treatment scans at liberal binarizing thresholds that returned to pre-scan levels on the following day. This decrease in network efficiency after treatment could have implications for understanding analgesic mechanisms and the effects of chronic pain on brain function.

Disclosures: **T. Bodily:** None. **J. Peacock:** None. **D. Busath:** None. **B. Kirwan:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.16/GG8

Topic: D.03. Somatosensation: Pain

Support: NRF/2016M3C7A1904984

Title: Decreased resting state network of fibromyalgia in theta band using graph filtration based on persistent homology

Authors: ***J. KIM**, M. CHOE, M. LIM, C. CHUNG
Seoul Nat Univ., Seoul, Korea, Republic of

Abstract: Fibromyalgia (FM), chronic widespread pain, exhibits spontaneous pain without external stimuli and is associated with alteration in intrinsic brain networks at rest. To understand the topological features of brain network in FM, we employed persistent homology which is a multiple scale network modeling framework without thresholding. Magnetoencephalography (MEG) activity was recorded in 19 healthy controls (HCs) and 18 FM patients at rest. The network features such as barcode, single linkage dendrogram and single linkage matrix were generated based on the proposed modeling framework in theta, alpha, beta, and gamma frequency. In theta band, the slope of decrease in barcodes showed steeper in HC, suggesting FM patients had decreased global connectivity. FM patients had reduced connectivity within default mode network, between middle temporal lobule and visual area, and between inferior temporal gyrus and cuneus in theta frequency. The longer pain duration was correlated with reduced connectivity between middle temporal lobule and visual cortex and between inferior temporal gyrus and visual cortex. Our results provided decreased intrinsic connectivity within default mode network and sensory network of FM patients in theta band. Our findings demonstrated that the aberrant resting state network would be associated with cognitive dysfunction and disrupted sensory processing without sensory stimuli in chronic pain. The persistent pain of FM may contribute to disruption of functional connectivity during resting state.

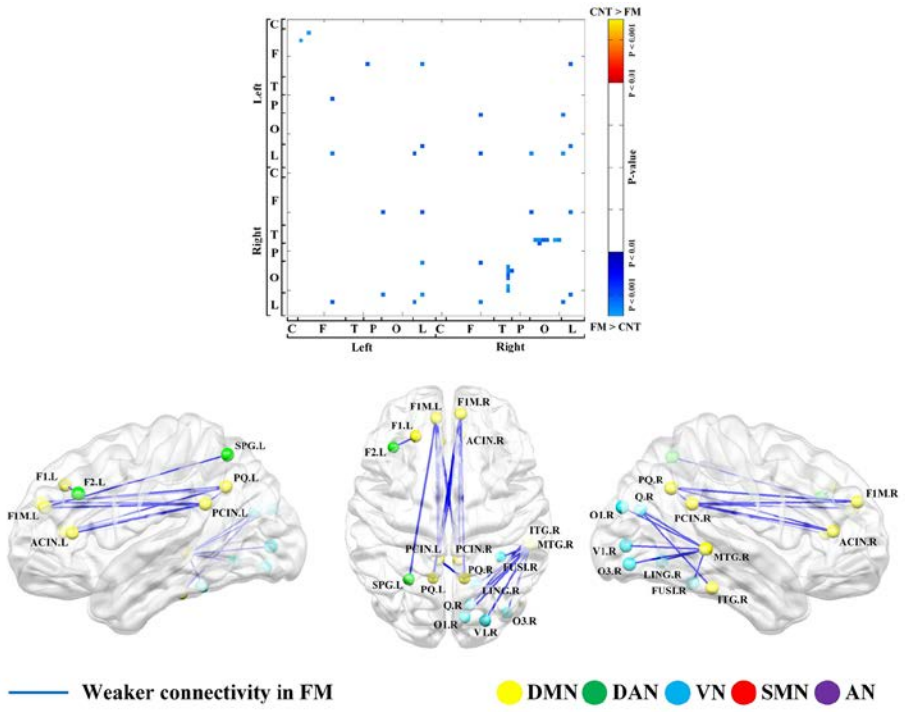


Figure 1. Local network properties based on persistent homology in theta band. Comparison of single linkage matrix (SLM) between fibromyalgia (FM) patients and healthy controls (HCs) (*upper*). Difference in the resting state network between FM and HC ($p < 0.001$) (*lower*).

Disclosures: **J. Kim:** None. **M. Choe:** None. **M. Lim:** None. **C. Chung:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.17/GG9

Topic: D.03. Somatosensation: Pain

Support: NIH/NCCIH (P01-AT006663)

Korean Institute of Oriental Medicine

Title: Predicting clinical pain states using cerebral blood flow in chronic low back pain: A machine learning approach

Authors: I. MAWLA[1], J. LEE[2], M. LOGGIA[2], A. ORTIZ[2], J. KIM[3], H. KIM[3], C. JUNG[3], S.-T. CHAN[2], N. MALEKI[2], J. GERBER[2], R. EDWARDS[4], A. WASAN[5], C. BERNA[6], J. KONG[2], T. KAPTCHUK[7], R. GOLLUB[2], B. ROSEN[2], *V. NAPADOW[2]
[2]Martinos Ctr. for Biomed. Imaging, [1]Massachusetts Gen. Hosp., Charlestown, MA; [3]Div. of

Clin. Res., Korea Inst. of Oriental Med., Yuseong-gu, Korea, Republic of; [4]Anesthesiology, Perioperative and Pain Med., Brigham and Women's Hosp., Chestnut Hill, MA; [5]Anesthesiol. and Pain Med., Univ. of Pittsburgh Med. Ctr., Pittsburgh, PA; [6]Univ. Hosp. of Lausanne, Lausanne, Switzerland; [7]Beth Israel Deaconess Med. Ctr., Boston, MA

Abstract: Prediction of pain states using brain imaging markers has been mostly limited to evoked experimental stimuli in healthy controls. Fewer efforts have been made towards the identification of brain-based predictive markers of different states in clinical pain. Chronic Low Back Pain (CLBP) is a good target to examine different states of clinical pain as back pain can be exacerbated by performing various physical maneuvers which evoke patients' clinical pain. In two independent studies with CLBP patients (total N=90), we used Arterial Spin Labeling (PCASL, 6 minutes, TR/TE=3800/15 ms, label duration=1500 ms, post label delay=1200 ms, at 3T) to characterize Regional Cerebral Blood Flow (RCBF) before and after a clinical back pain exacerbation procedure through painful maneuvers (Wasan, Loggia, et al., 2011). PCASL images were preprocessed as follows: motion correction, skull-stripping, tag-control subtraction, RCBF estimation in absolute values (mm/100 g of tissue/min), registration to T1 image, whole-brain RCBF normalization, non-linear transformation to common space (MNI152), and further spatial smoothing (FWHM=8 mm). Maneuvers successfully increased back pain intensity (pre-maneuver=29.4, SD=20.6, post- maneuver=50.5, SD=20.7, $p<0.0001$, on a 0-100 scale). Univariate GLM analysis (combined usable data from studies 1 & 2, N=78) showed that painful maneuvers increased RCBF in the thalamus, putamen, caudate, amygdala, hippocampus, posterior cingulate, supplementary motor area (SMA), preSMA, medial prefrontal (MPFC), pregenual anterior cingulate, inferior frontal gyrus (IFG) and ventrolateral prefrontal (VLPFC) (cluster-corrected, $Z=2.3$, $P<0.05$). Furthermore, in study 1 (N=55, training set), multivariate classification analysis (support vector machine with linear kernel) was applied to discriminate the low-pain (pre-maneuver) and high- pain (post-maneuver) state from RCBF data. Significant positive classifier weights were found in the thalamus, putamen, SMA, paracentral lobule, primary motor cortex (M1), IFG, MPFC, VLPFC, and mid-Insula. The accuracy of the classification in the training set was 70% (sensitivity=72.2%, specificity=67.3%, precision=69.0%, AUC=0.74). The classifier weight map calculated in study 1 was then tested in the independent testing dataset (study 2, N=23), and led to similar prediction results, with 73.9% accuracy (sensitivity=82.6%, specificity=65.2%, precision=70.4%, AUC=0.78). These results demonstrate the potential utility of multivariate machine learning based on cerebral blood flow information to predict clinical pain states.

Disclosures: I. Mawla: None. **J. Lee:** None. **M. Loggia:** None. **A. Ortiz:** None. **J. Kim:** None. **H. Kim:** None. **C. Jung:** None. **S. Chan:** None. **N. Maleki:** None. **J. Gerber:** None. **R. Edwards:** None. **A. Wasan:** None. **C. Berna:** None. **J. Kong:** None. **T. Kaptchuk:** None. **R. Gollub:** None. **B. Rosen:** None. **V. Napadow:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.18/GG10

Topic: D.03. Somatosensation: Pain

Support: Ministry of Trade, Industry & Energy: 10073159

Title: Neurometabolite changes in patients with complex regional pain syndrome: A magnetic resonance spectroscopy study

Authors: *Y.-H. JUNG[1], H.-J. KIM[2], S. JEON[2], J. KWON[2], Y. KIM[2], D.-H. KANG[2]
[1]Dept. of Neuropsychiatry, [2]Seoul Natl. Univ. Hosp., Seoul, Korea, Republic of

Abstract: The first aim was to investigate distinct neurometabolites in the right and left thalamus and insula of complex regional pain syndrome (CRPS) patients using proton magnetic resonance spectroscopy (MRS). Levels of N-acetylaspartate (NAA), N-acetylaspartylglutamate (NAAG), myo-inositol (mI), glutamine (Gln), glycerophosphocholine (GPC), glutathione (GSH), and alanine (Ala) relative to total creatine (tCr) levels were determined in 12 CRPS patients compared to 11 healthy controls using MRS. Levels of NAAG/tCr and Ala/tCr were higher in CRPS patients than in controls in the left thalamus. NAAG/tCr, mI/tCr, and Gln/tCr levels were higher, but NAA/tCr levels were lower in the right insula of CRPS patients. There were negative correlations between GSH/tCr and pain score in the left thalamus. The second aim was to assess neurometabolites affecting neuroinflammation in CRPS patients using [^{11}C]-(R)-PK11195 positron emission tomography (PET) and MRS. We found positive correlations of Lip13a/tCr and Lip09/tCr with the DVR of [^{11}C]-(R)-PK11195 in CRPS patients. There were positive correlations between plasma C-reactive protein (CRP) and GPC/tCr in the right thalamus in CRPS patients. There were positive correlations between CRP and Ala/Glx (Glu+Gln), GPC/Glx and mI/Glx, but negative correlations between CRP and Glx/tCr in the left thalamus in CRPS patients. There was negative correlation between CRP and mI/tCr in the left insula in CRPS patients. The third aim was to investigate whether distinct neurometabolites would affect depression, anxiety, suicidal ideation and anger in CRPS patients. In the right thalamus, there were positive correlations between Val/tNAA (NAA + NAAG) and the BAI and between GPC/Lip13a and the SSI and a negative correlation between Gln/NAA and the BDI. In the left thalamus, there were positive correlations between Ala/Gln and the BDI and SSI, between Glu/Gln and the BDI and SSI, between NAAG/Gln and the BDI, and between Ala/Lip13a and the BAI and a negative correlation between Gln and the SSI. In the right insula, there was a positive correlation between Ala/NAAG and the STAXI-T (trait anger) and a negative correlation between Cr/NAA and the STAXI-T. In the left insula, there was a negative correlation between Cr/Glx and the STAXI-T. The complex pathophysiological mechanisms

underlying CRPS include the involvement of the central nervous system, parasympathetic nervous system, neuroinflammation, oxidative stress, and antioxidants. Thus, the distinct metabolites may be essential to understand the core pathophysiology of CRPS, and combinatorial biomarkers have a strong diagnostic and prognostic potential for CRPS.

Disclosures: Y. Jung: None. **H. Kim:** None. **S. Jeon:** None. **J. Kwon:** None. **Y. Kim:** None. **D. Kang:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.19/GG11

Topic: D.03. Somatosensation: Pain

Support: NCCIH Grant R21-AT007247

NCCIH Grant F32-AT006949

NCCIH Grant K99-AT008238

NCCIH Grant F30-AT009165

NINDS Grant R01-NS239426

Mind and Life Institute Francisco J. Varela Award

Wake Forest Center for Integrative Medicine

Title: Brain mechanisms moderating the relationship between depression and pain

Authors: *A. ADLER-NEAL[1], N. EMERSON[3], S. FARRIS[1], Y. JUNG[2], J. G. MCHAFFIE[1], R. C. COGHILL[4], F. ZEIDAN[1]
[1]Neurobio. and Anat., [2]Dept. of Radiology, Wake Forest Sch. of Med., Winston Salem, NC; [3]Neurosurg., Johns Hopkins Med., Baltimore, MD; [4]Cincinnati Children's Hosp., Cincinnati, OH

Abstract: Rationale:
Pain and depression exhibit a co-morbid relationship. Depression is associated with heightened activation in the prefrontal (PFC) and insular cortices, brain regions associated with facilitating affective and sensory-discrimination of pain, respectively. However, the neural mechanisms moderating the relationship between depression and pain are unknown.
Methods: Seventy-six healthy, pain-free volunteers completed the Beck Depression Inventory (BDI) and fell below the cutoffs for clinically significant depression. Pseudocontinuous arterial spin labeling (PCASL) was used to acquire whole-brain cerebral blood flow images. Noxious

thermal stimuli were delivered to the right calf across four PCASL scans. Pain intensity and unpleasantness visual analog scale ratings (VAS) were collected after each scan. Heat scans consisted of alternating patterns of 49°C and 35°C. Neutral scans contained only 35°C stimuli. Separate regression analyses tested the relationship between BDI and pain intensity and unpleasantness ratings (SPSS 19). First and second-level repeated-measures ANOVAs [The Functional Magnetic Resonance Imaging of the Brain (FMRIB) Software Library] examined the main effect of pain (heat vs. neutral series) within and across individuals, respectively. Demeaned BDI scores, pain intensity, and pain unpleasantness ratings were entered as the first, second, and third regressors, respectively. The interaction between BDI and a) pain intensity and b) pain unpleasantness was modeled as the fourth and fifth regressor, respectively. Post hoc region of interest analyses examined significant interactions.

Results:

Greater BDI scores were associated with greater pain intensity (r=. 32; p=.006) and unpleasantness (r=.34; p=.003) ratings. Heat series produced greater pain-related activation in the primary somatosensory cortex (SI) corresponding to the stimulation site, thalamus, cerebellum, secondary somatosensory cortex (SII), and anterior/posterior insula, when compared to neutral series. Greater ventrolateral PFC (p=.006), anterior insula (p=.03), SII and posterior insula (p=.01) activation moderated the positive relationship between depression and pain intensity. Greater SI activation corresponding to the stimulation site (p=.014) and decreased orbitofrontal cortex (p=.003), anterior insula (p=.04), and rostral anterior cingulate cortex (p<.05) activation moderated the positive relationship between depression and pain unpleasantness. These results provide novel evidence that functionally distinct brain processes moderate the relationship between depression and pain intensity and unpleasantness.

Disclosures: A. Adler-Neal: None. **N. Emerson:** None. **S. Farris:** None. **Y. Jung:** None. **J.G. McHaffie:** None. **R.C. Coghill:** None. **F. Zeidan:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.20/GG12

Topic: D.03. Somatosensation: Pain

Support: BNI Semilla Grant

OAIC Grant 694/14

Soc. Anest. Chile Grant 2016

Title: Effects of opioid-induced analgesia on electroencephalographic markers of pain perception

Authors: *J. I. EGANA[1,2], R. MONTEFUSCO-SIEGMUND[2], A. BLANCH[1], D. ROJAS-LIBANO[3], G. RIVERA[4]
[1]Anestesiología y Reanimación, Hosp. Clinico Univ. De Chile, Santiago, Chile; [2]Biomed. Neurosci. Inst., Santiago, Chile; [3]Facultad de Educación, Univ. Alberto Hurtado, Santiago, Chile; [4]Dept. de Kinesiología, Univ. De Chile, Santiago, Chile

Abstract: Chronic pain is a highly prevalent condition in modern medicine. Is the main cause of consultation, the principal cause of disability worldwide and represent enormous costs for all health systems. In most cases, chronic pain is a consequence of long exposures to untreated or subtreated acute pain. Efforts should be done in situation where acute pain is expected in order to prevent its transformation into chronic pain. Surgery is one of those situations. Nevertheless, chronic post-surgical pain (CPSP) is highly prevalent indicating that surgery-derived acute pain is not being correctly detected and/or treated. Mechanisms that account for acute to chronic pain transformation start acting rapidly after intense painful/nociceptive stimulation. That makes necessary to maintain adequate levels of analgesia during the surgery itself. Currently, there are no reliable intraoperative pain/nociception monitors. EEG is a powerful tool that has proved its utility for monitoring the brain under general anesthesia. Unfortunately most of the achievements have been developed in the study of unconsciousness and amnesia with little progress in the pain/nociception field. Before the EEG can guide medical interventions, the electrical activity associated with pain and analgesia should properly be characterized. Eight subjects were submitted to a 2 stages experiment while a 16+8 electrodes EEG was recorded. In the first stage they underwent a protocol of painful transcutaneous electrical stimulation of variable intensity. 3 consecutive stimuli of 4 different intensities, ranging from mild to moderate pain, were applied. Subject were instructed to report the amount on pain elicited by each of the stimuli in a 0-10 scale. During the second stage subjects repeated the previous protocol while they were exposed to increasing levels of analgesia. Analgesia was obtained by intravenous computer-controlled injection of remifentanil. Remifentanil is an ultrashort-acting synthetic opioid that allow quick effect and rapid recovery allowing rapid changes in analgesic effect. We report the effects of opioid-induced analgesia on pain-associated Event Related Potentials (pERP) and on induced oscillatory activity.

Disclosures: J.I. Egana: None. **R. Montefusco-Siegmund:** None. **A. Blanch:** None. **D. Rojas-Libano:** None. **G. Rivera:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.21/GG13

Topic: D.03. Somatosensation: Pain

Support: NIH Grant R01DE022746

Title: Velocity of brain-state trajectory encodes salience and attention rather than pain

Authors: *R. JABAKHANJI[1], A. T. BARIA[1], M. A. FARMER[1], A. V. APKARIAN[1,2,3]
[1]Dept. of Physiol., [2]Dept. of Anesthesiol., [3]Dept. of Physical Med. and Rehabil., Northwestern Univ. Feinberg Sch. of Med., Chicago, IL

Abstract: Distinct perceptions are linked to distinct spatio-temporal patterns of activity. Different odors, for example, have been shown to elicit unique and reliable spatio-temporal activity patterns, or trajectories, which are thought to reflect not only stimulus properties, but also the subjective experience of stimuli. While distinct activity trajectories have been demonstrated in visual and olfactory systems, little is known about how these trajectories manifest in pain. Here we investigated the dynamics of fMRI brain activity trajectories in healthy subjects as they rated or passively underwent various painful and non-painful stimuli.
Fourteen subjects received block stimulation of painful thermal and visual stimuli in separate scans. Thirteen subjects received block stimulation of painful and non-painful vulvar touch in separate scans. Finally, 6 subjects received a thermal stimulus designed to elicit varying magnitudes of pain while the stimulus was held constant. Additionally, for a smaller subset of these scans, painful stimuli were passively experienced, and not rated. Each scan consisted of 6-12 stimulus blocks, each block lasting between 15-30 seconds. Functional images were preprocessed by ICA de-noising, regressing out signal related to motion, physiological artifact, white-matter, and average global activity. BOLD pattern dynamics related to each stimulus were then evaluated in activity space composed of the time series of these 480 regions, in which the location of a single data point represented the spatial activity pattern at a single point in time, and all data points temporally ordered represented the trajectory of brain activity. Thus, greater distance between 2 data points in this space indicated a greater dissimilarity in the brain activity patterns. Likewise, the distance between temporally adjacent data points indicated the velocity of the spatial pattern, or how quickly activity traversed along the trajectory. We observed primarily how the velocity of brain activity differed between the different thermal, somatosensory, and visual stimuli.
We found that activity velocity was slowed 1) in thermal painful compared to visual stimuli, 2) when pain was abolished under normally painful thermal stimuli, and 3) for both painful and non-painful vulvar touch. We also found that rating stimuli as they occurred further slowed activity velocity, and resulted in highly different activity patterns. These results indicate that fMRI activity velocity more likely encodes salience and attention, rather than perception of pain or stimulus properties alone. However, further analysis remains to be done on other activity trajectory features.

Disclosures: R. Jabakhanji: None. **A.T. Baria:** None. **M.A. Farmer:** None. **A.V. Apkarian:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.22/GG14

Topic: D.03. Somatosensation: Pain

Support: "Brain machine Interface Development" under the Strategic Research Program for Brain Sciences supported by the Japan Agency for Medical Research and Development (AMED)

National Institute for Information and Communications Technology, Japan

Wellcome Trust, UK

the W.D. Armstrong Fund, UK

the Cambridge Trust, UK

Title: Closed-loop pain relief control using fMRI multi-voxel decoder and reinforcement learning

Authors: *S. ZHANG[1], H. MANO[2], W. YOSHIDA[3], M. KAWATO[3], T. YANAGISAWA[4], K. SHIBATA[5], B. SEYMOUR[1,2,3]
[1]Dept. of Engin., Univ. of Cambridge, Cambridge, United Kingdom; [2]Ctr. for Information and Neural Networks, NICT, Osaka, Japan; [3]ATR, Kyoto, Japan; [4]Osaka Univ., Osaka, Japan; [5]Nagoya Univ., Nagoya, Japan

Abstract: A major objective of technology-based therapeutics for chronic pain is to construct a smart 'closed-loop' automatic control system that is capable of adjusting its intervention according to brain activity related to pain intensity experienced. We demonstrated in our study the feasibility of such system in healthy human participants using fMRI, and explored the potential of brain-machine co-adaptation.
Firstly, in the decoder construction session, individual participant's functional brain images were recorded during fMRI scanning for decoder training, during which high and low levels of painful electrical stimuli were delivered in a sequence of random trials to elicit two levels of pain. Individual-specific, multi-voxel decoder was then trained for automatic classification of pain level experienced, using regions of interest identified in previous pain studies. Secondly, in the adaptive control session, the intensity of pain stimuli delivered was controlled by the participant's decoded pain from their brain activity in the previous trial. Specifically, after receiving pain, the decoder estimated in real-time the participants' probability of experiencing high pain from their brain images, which was used to determine the pain intensity level to be delivered in the next trial. To complete the control loop, the pain delivery system learned to

lower the overall level of pain delivered to the participant, which was accomplished with a basic reinforcement learning algorithm by rewarding the stimulation state that elicited lower decoded pain signal in the participant. Trial structure was otherwise identical to that of the decoder construction session, which made it a yoked control condition to the adaptive control session effectively, allowing investigation of whether any brain-machine co-adaptation processes took place.
Pilot results showed that such closed-loop system was achievable from an engineering perspective, and the pain delivery system could learn quickly to deliver low pain to participants, despite the probabilistic nature of the decoder. Despite the limited temporal resolution of fMRI scanning, the current system nevertheless demonstrates the use of machine learning techniques based on brain activity can be adopted in principle for closed-loop pain control systems.

Disclosures: S. Zhang: None. **H. Mano:** None. **W. Yoshida:** None. **M. Kawato:** None. **T. Yanagisawa:** None. **K. Shibata:** None. **B. Seymour:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.23/GG15

Topic: D.03. Somatosensation: Pain

Support: R01 AT007176

Title: Pathological functional connectivity and pain catastrophizing converge on the salience and cingulo-opercular networks

Authors: *S. KRIMMEL[1], M. KEASER[2], J. HAYTHORNTHWAITE[3], D. A. SEMINOWICZ[4]
[1]Neural and Pain Sci., [2]Univ. of Maryland Baltimore, Baltimore, MD; [3]Johns Hopkins Univ., Baltimore, MD; [4]Dept of Neural & Pain Sci., Univ. of Maryland, Baltimore, Baltimore, MD

Abstract: Migraine is a chronic pain disorder with substantial disease burden. A hallmark of chronic pain disorders is high levels of pain catastrophizing, which is associated with disease burden and maintenance. We acquired resting state functional MRI (rsfMRI) data in 21 episodic migraine patients (4-14 headache days per month) and 21 healthy controls matched on age, sex, race, education, and BMI. We determined resting state functional connectivity (RSFC) by extracting timeseries data from 264 nodes and used the extracted timeseries to create a full correlation matrix (264 x 264). To elucidate which functional connections and brain regions may promote pain catastrophizing, we correlated Pain Catastrophizing Scale (PCS) scores to the RSFC matrix ($p<0.05$, false discovery rate corrected for multiple comparisons). Consistent with existing literature on pain catastrophizing, we found migraine patients displayed higher PCS scores than controls. Additionally, we found PCS in patients correlated with RSFC in nearly all

brain networks, particularly to salience, cingulo-opercular, and default mode networks. Also, we identified differences in RSFC between patients and controls that was localized primarily to the salience, cingulo-opercular, and sensory networks. An overlap of these significant findings shows that the salience network and cingulo-opercular networks display both differences in RSFC between patients and controls and correlate to patient PCS scores. These findings support attention based theories of pain catastrophizing in chronic pain.

Disclosures: S. Krimmel: None. **M. Keaser:** None. **J. Haythornthwaite:** None. **D.A. Seminowicz:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.24/GG16

Topic: D.03. Somatosensation: Pain

Support: R61AT009310

R01AT006364

R01AT008563

P01 AT006663

R21AT008707

Title: Brain responses evoked by acupuncture and imagery

Authors: *J. CAO[1,2], J. PARK[1], K. JORGENSON[1], C. LANG[1], J. LIU[1], R. GOLLUB[1], S. ORR[1], J. KONG[1]
[1]PSYCHIATRY, MASSACHUSETTS GENERAL HOSPITAL, Charlestown, MA; [2]Insitution of Acupuncture moxibustion and tuina, Beijing Univ. of Chinese Med., Beijing, China

Abstract: Introduction: The use of imagery to treat illnesses is a widely accepted medical practice that has been used for long time, but the underlying mechanism behind its efficacy remains unknown. Acupuncture is a unique, invasive treatment modality. Studies have shown that the experience[1], as well as the visualization[2], of acupuncture needle stimulation provokes activation in overlapping brain regions such as the anterior insula (AI) and periaqueductal grey (PAG). In this study, we took advantage of the unique characteristic of acupuncture, and investigated fMRI signal changes evoked by acupuncture and video-guided acupuncture imagery treatment (VGAIT).
Methods: To date, 10 right-handed, healthy individuals have completed the crossover-designed

study. We investigated fMRI signal changes evoked by 1) VGAIT (watching a video of an acupuncture needle stimulating subject's acupoint, and imagining it being applied concurrently), 2) a VGAIT control condition (watching a video of a cotton swab touching subject's bodies and imagining it being applied concurrently), 3) real acupuncture, and 4) sham acupuncture (Streitberger needle). fMRI data were collected using a 3.0 T MRI scanner while subjects completed two 9-minute fMRI scans during which intermittent acupuncture, VGAIT and corresponding control conditions were applied. Two acupoints (right SP 6 and SP 9) were tested in this study. Data analysis was performed using SPM 12 with a threshold of $p< 0.005$ over 40 continuous voxels.

Results: Compared with the VGAIT control group, the VGAIT group showed fMRI signal increases at the bilateral dorsolateral prefrontal cortex (DLPFC), cerebellum, right PAG and temporoparietal junction. No significant fMRI signal decreases were observed. Compared with needle-stimulated acupuncture, VGAIT produced fMRI signal increases in the bilateral occipital cortex, hippocampus, PAG, pons, and cerebellum, and fMRI signal decreases in the right superior parietal lobule and left striatum. Actual acupuncture produced significant fMRI signal increases in the right superior temporal gyrus, dorsal lateral prefrontal cortex, and left posterior cingulate cortex compared with sham acupuncture.

Conclusions: Our results suggest that VGAIT can activate a widespread brain network, including PAG, a key region in the descending pain modulation system. This finding demonstrates the potential of VGAIT in pain management.

References:

1.Kong, J., et al., *Test-retest study of fMRI signal change evoked by electroacupuncture stimulation.* Neuroimage, 2007. **34**: 1171-81.

2.Cheng, Y., et al., *Expertise modulates the perception of pain in others.* Curr Biol, 2007. **17**: 1708-13.

Disclosures: J. Cao: None. **J. Park:** None. **K. Jorgenson:** None. **C. Lang:** None. **J. Liu:** None. **R. Gollub:** None. **S. Orr:** None. **J. Kong:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.25/GG17

Topic: D.03. Somatosensation: Pain

Support: Multiple Sclerosis Society of Canada

CIHR

Mayday Fund

Title: Altered resting state oscillatory MEG power in multiple sclerosis patients with chronic pain

Authors: *J. A. KIM[1,2], R. BOSMA[1], K. S. HEMINGTON[1,2], A. ROGACHOV[1,2], J. C. CHENG[1,2], N. R. OSBORNE[1,2], J. OH[3,2], K. D. DAVIS[1,2]
[1]Krembil Res. Inst., Toronto, ON, Canada; [2]Univ. of Toronto, Toronto, ON, Canada; [3]St. Michael's Hosp., Toronto, ON, Canada

Abstract: Objectives:
The mechanisms underlying pain associated with multiple sclerosis (MS) are not completely understood despite a high prevalence of patients whose quality of life suffers from the presence of chronic pain. In general, magnetoencephalography (MEG) and electroencephalogram studies have reported that MS patients have increased slow wave power in the theta and delta bands (Leocani et al 2000) and reduced alpha band coherence (Cover et al. 2006) compared to healthy controls. The latter findings suggest an alpha band-related abnormality in brain communication in MS. However, it is not known how chronic pain in MS impacts oscillatory brain activity, as measured using MEG. MRI studies have indicated that chronic pain is associated with structural and functional abnormalities in the dynamic pain connectome (Kucyi and Davis 2015; Bosma et al. 2017), a system that includes the default mode network, salience network, ascending nociceptive and descending modulation pathways. The goal of the current study was to use MEG to determine whether resting state brain oscillatory power is abnormal in the dynamic pain connectome in patients with MS and if this is related to chronic pain and its impact on quality of life metrics.
Methods:
Patients with MS and age-/sex-matched healthy control (HC) subjects were recruited for the study that was approved by the local institutional research ethics board and all participants provided informed written consent to the study protocol. Every participant underwent a 5 min MEG resting state data acquisition, a comprehensive psychophysics protocol, and completed questionnaires to assess pain, personality, clinical, psychological and quality of life issues. The brain power spectra information was derived from MEG data using linearly-constrained minimum variance beamformed time series of the resting state scan.
Results:
Whole brain power spectra in MS patients and HCs were similar over most frequency bands except the alpha band. Furthermore, the MS patients had altered alpha band power in specific nodes of the dynamic pain connectome that was related to their chronic pain and reductions in quality of life metrics that assessed the impact of pain (e.g., pain interference from the Brief Pain Inventory).
Conclusions:
These findings indicate that MS chronic pain affects resting state brain activity at slow-medium frequencies. This suggests that patients with chronic pain have altered baseline levels of activity which can lead to the manifestation of pain-related behaviour.

Disclosures: J.A. Kim: None. **R. Bosma:** None. **K.S. Hemington:** None. **A. Rogachov:** None. **J.C. Cheng:** None. **N.R. Osborne:** None. **J. Oh:** None. **K.D. Davis:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.26/GG18

Topic: D.03. Somatosensation: Pain

Support: NIH (HL117664, NS096761, EB021027, AT009263, S10OD021721)

NSF DGE-1069104

Title: Quantification of tonic thermal pain using EEG data and random forest models

Authors: *V. VIJAYAKUMAR[1], M. CASE[2], S. SHIRINPOUR[2], B. HE[2]
[1]Electrical Engin., [2]Univ. of Minnesota, Minneapolis, MN

Abstract: Pain is considered to be the product of a conscious, multi-dimensional response to stimuli perceived as nociceptive in nature. The current standard for pain assessment is self-report, which shows high degree of variability in chronic pain patients and can lead to sub-optimal treatment methods. Hence, developing an accurate pain assessment system which is generalizable across populations can increase the efficacy of clinical treatments. The aim of this study is to use electroencephalography (EEG) data collected from a subject pool in response to tonic thermal pain to build a classification model for predicting the pain score in the range of 1-10 in an unseen subject from the subject pool. The contribution of each frequency band to model accuracy is analyzed. Source imaging is then used to analyze the spatial correlates of two simultaneous temporal responses observed in a subject, which are quantifiably the best indicators of nociception: One which is shared by all subjects in the subject pool, referred to as a global pain signature, and one which is unique to each subject, referred to as a local pain signature. Twenty-five healthy subjects were recruited for this study. The threshold and tolerance levels of each subject were measured. During the experiment tonic low, medium, and high thermal pain stimuli between threshold and tolerance were administered to the subject. To rate their pain, they used a custom-made continuous pain rating device on a color bar. A random forest model is trained to predict pain scores using time-frequency wavelet representations of independent components obtained from the EEG data. Principal Component Analysis (PCA) is used to extract the spatial maps of the most consistent electrode activity, and sLORETA is used for source localization on these components.
The mean classification accuracy for predicting pain on an unseen test subject for a range of 1-10 is 89.45% using the leave-one-out classification paradigm, which is 6% higher than existing state of the art for multi-class pain prediction. The gamma band is the most important to both inter-subject and intra-subject classification accuracy. Global correlates of pain mostly localized to the posterior parietal lobe while local correlates predominantly localized to the anterior cingulate cortex, showing that there are observable spatial differences between two concurrent temporal

pain responses in a subject. Our results demonstrate the potential of this tool to be used clinically to help improve chronic pain treatment, and establish spatial biomarkers for future pain-related studies using EEG.

Disclosures: V. Vijayakumar: None. **M. Case:** None. **S. Shirinpour:** None. **B. He:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.27/GG19

Topic: D.03. Somatosensation: Pain

Title: Functional and structural plasticity in meditators with chronic pain

Authors: *L. TULIPANI[1], M. SHPANER[2], J. BISHOP[2], N. LA ROSA[2], S. YOUNG[2], M. NAYLOR[2]
[1]Dept. of Psychiatry/Neuroscience Grad. Program, [2]Dept. of Psychiatry, Univ. of Vermont, Burlington, VT

Abstract: Mindfulness meditation can be helpful in coping with chronic pain. Subjective accounts of strong analgesia in experienced meditators are supported by recent behavioral and neuroimaging findings. The process of pain chronification is sustained by widespread changes in the central nervous system. However, the way that long-term meditation practice can affect the brain in chronic pain patients has not been investigated. In this cross-sectional study, we used neuroimaging to examine the neuroplasticity of coping with pain in chronic pain patients who are experienced meditators. Mindfulness practitioners with chronic pain (at least 1000 hours of practice) and meditation-naïve patients with chronic pain were matched for age and gender; all participants completed an acute pain task (n=13 for each group) as well as structural diffusion tensor imaging scans (n=18 meditators and n=21 naïve patients). Prior to scanning, we determined a moderate pain level for each participant (7 on a 10-point scale). The acute pain task consisted of two runs using induced thermal pain, with five 20-second pain blocks each. Participants were instructed to “attend to the pain” in the first run and to “decrease and control the pain” (cope) in the second run. Whole-brain analyses revealed significant between-group differences in three regions (dorsal Anterior Cingulate Cortex, left Inferior Frontal Gyrus and the cerebellum) during coping with pain when compared to attending to pain. These findings are consistent with existing literature on acute pain perception in experienced meditators, which report decreased activations within sensory-discriminative regions of the pain matrix with increased meditation experience. Whole-brain analyses of axial diffusivity (AD) revealed higher AD in the superior and inferior longitudinal fasciculi in the meditation-naïve patients, possibly indicating stronger corticolimbic connections in the absence of meditation experience.

Disclosures: **L. Tulipani:** None. **M. Shpaner:** None. **J. Bishop:** None. **N. La Rosa:** None. **S. Young:** None. **M. Naylor:** None.

Poster

398. Pain Imaging

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 398.28/GG20

Topic: D.03. Somatosensation: Pain

Support: NIH Grant R61AT009306

The Research Council of Norway/European Union's Seventh Framework Programme for research, technological development and demonstration Grant 240553/F20

Title: Patient-clinician concordance in social mirroring brain circuitry during pain treatment: A hyperscanning fMRI study

Authors: *D.-M. ELLINGSEN[1], C. JUNG[1], K. ISENBURG[1], J. GERBER[1], I. MAWLA[1], R. SCLOCCO[1], R. R. EDWARDS[2], J. KELLEY[3], I. KIRSCH[4], T. J. KAPTCHUK[4], V. NAPADOW[1]
[1]Martinos Ctr. for Biomed. Imaging, Massachusetts Gen. Hospital, Harvard Med. Sc, Charlestown, MA; [2]Brigham and Women's Hosp., Boston, MA; [3]Endicott Col., Beverly, MA; [4]Harvard Med. Sch., Boston, MA

Abstract: The patient-clinician relationship can modulate symptoms such as pain, but the brain basis for this is unknown. We simultaneously recorded functional Magnetic Resonance Imaging (fMRI hyperscanning) in patients and clinicians, who interact via video transfer, during clinician-initiated treatment of the patient's pain. We hypothesized concordant activation of circuitry involved in social mirroring, such as ventrolateral Prefrontal Cortex (vlPFC) and anterior Insula (aINS) in both patients and clinicians during pain treatment. Patients with chronic pain (fibromyalgia) and clinicians (acupunturists) were enrolled. Each patient was matched with a clinician participant (12 patient-clinician 'dyads'). The patient received a number of moderately painful cuff pressures to the left leg (15 s), while the clinician used a button box to control (real or sham) electroacupuncture stimulation to the patient's leg to treat the cuff pain. Both participants used a Visual Analogue Scale to rate self-pain (patients) or vicarious pain (clinicians) after each trial. Using MRI-compatible cameras, the participants were enabled to communicate non-verbally throughout the scan. fMRI preprocessing included motion correction, skull stripping, correction for magnetic field inhomogeneities, and registration to MNI152. After single-subject GLM analysis, we conducted a group GLM (whole-brain cluster-correction for multiple comparisons) followed by a group conjunction analysis, between patients and clinicians, of the [pain,treatment]>[rest] contrast. Patients reported decreased pain for both real and sham

treatment, compared to overt no-treatment, which was echoed by reduced ratings of vicarious pain by the clinicians. Patients (receiving pain+treatment) showed activation of posterior (pINS) and mid-anterior (m-aINS) Insula, secondary somatosensory areas (SII), Supplementary Motor Area (SMA), ventrolateral Prefrontal Cortex (vlPFC) dorsolateral prefrontal cortex (dlPFC), and Temporoparietal Junction (TPJ). Clinicians (observing pain+treating) showed activation in aINS, vlPFC, dorsomedial PFC, dlPFC, TPJ, Superior Temporal Sulcus (STS), and primary and secondary visual areas. A conjunction analysis showed activation of vlPFC, aINS, and TPJ for both patients and clinicians during pain and treatment, indicating concordant activation of circuitry important for social mirroring in interacting patients and clinicians.

Disclosures: D. Ellingsen: None. **C. Jung:** None. **K. Isenburg:** None. **J. Gerber:** None. **I. Mawla:** None. **R. Sclocco:** None. **R.R. Edwards:** None. **J. Kelley:** None. **I. Kirsch:** None. **T.J. Kaptchuk:** None. **V. Napadow:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.01/GG21

Topic: D.06. Audition

Support: EU H2020 FET Flagship: Human Brain Project (grant no. 720270)

Title: Expanding the Waxholm Space rat brain reference atlas using a data enriched magnetic resonance imaging template: New delineations of the auditory system, thalamus, and more

Authors: J. IMAD[1], A. E. WENNBERG[1], K. K. OSEN[1], F. CLASCÁ[2], G. CSUCS[1], C. COELLO[1], J. G. BJAALIE[3], *T. B. LEERGAARD[1]
[1]Inst. of Basic Med. Sci., Univ. of Oslo, Oslo, Norway; [2]Histology and Neurosci., Autonomous Univ. of Madrid, Madrid, Spain; [3]Univ. Oslo, Oslo, Norway

Abstract: Brain reference atlases are important resources for assigning anatomical location to experimental data, and for planning and interpreting experimental results. Three-dimensional atlases can also serve as templates for spatial co-registration (integration) and comparison of different types of brain images. The Waxholm Space atlas of the rat brain is a public resource based on high resolution magnetic resonance imaging (MRI) and diffusion tensor imaging (DTI) data. It features a coordinate system based on internal landmarks (Waxholm Space) and delineations of so far 79 brain structures (Papp et al., Neuroimage 97:374-386, 2015; Kjonigsen et al., Neuroimage 108:441-449, 2015). The atlas is shared via the Neuroimaging Informatics Tools and Resources Clearinghouse (www.nitrc.org), and has since its release been widely adopted by the community. A limitation is, however, that detailed anatomical delineations of several major brain regions are missing. In some instances, delineations are not easily introduced

due to low contrast in the underlying MRI / DTI templates, or complex features that are challenging to interpret in MRI / DTI data. To amend this, we have spatially registered images of serial histological sections stained for cyto-, chemo- and myeloarchitecture to the Waxholm Space rat brain template, and used these to aid the interpretation of boundaries of structures. We here outline our approach to defining brain structures in the Waxholm Space rat brain template, and present new delineations of the ascending auditory system, the thalamus, as well as several regions in the cerebral cortex and basal forebrain. The anatomical criteria underlying the delineations will be published, and the next version of the atlas delineations will be shared from www.nitrc.org.

Disclosures: J. Imad: None. **A.E. Wennberg:** None. **K.K. Osen:** None. **F. Clascá:** None. **G. Csucs:** None. **C. Coello:** None. **J.G. Bjaalie:** None. **T.B. Leergaard:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.02/GG22

Topic: D.06. Audition

Support: BBSRC BB/M00905X/1

Title: Cell type-specific and age-related changes in auditory cortical processing

Authors: D. LYNGHOLM, *S. SAKATA
SIPBS, Univ. of Strathclyde, Glasgow, United Kingdom

Abstract: Aging is inevitable and while the age-related deterioration of many brain functions including sensory, cognitive and motor functions are well documented, we know little about age-related changes in cortical processing at the neural circuit level. In particular, it remains unclear how cortical GABAergic interneurons contribute to age-related changes in cortical information processing. To address this issue, it is important to not only discriminate cell types, but also to delineate between central and peripheral effects of aging. In the present study, focusing on the auditory cortex, we investigate how the structure of spontaneous and auditory-evoked population activity changes with age and comparing two mouse strains to distinguish central and peripheral aging effects. We recorded neural ensembles in the auditory cortex of awake C57BL/6J mice, which have a mutation leading to early peripheral age-related hearing loss (ARHL). We observed an age-related decrease in multi-unit spiking activity, with a correlation between increased hearing threshold and increased spontaneous theta oscillations that is not apparent in age-matched CBA/J mice, which do not exhibit early ARHL. Combining electrophysiological and optogenetic approaches in both young and aged mice with and without early ARHL, we further investigate both cell-autonomous and circuit level changes in auditory cortical processing

involving GABAergic cells expressing either Parvalbumin or Somatostatin, thereby providing a deeper insight into the underlying mechanisms of age-related changes in auditory function.

Disclosures: D. Lyngholm: None. **S. Sakata:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.03/GG23

Topic: D.06. Audition

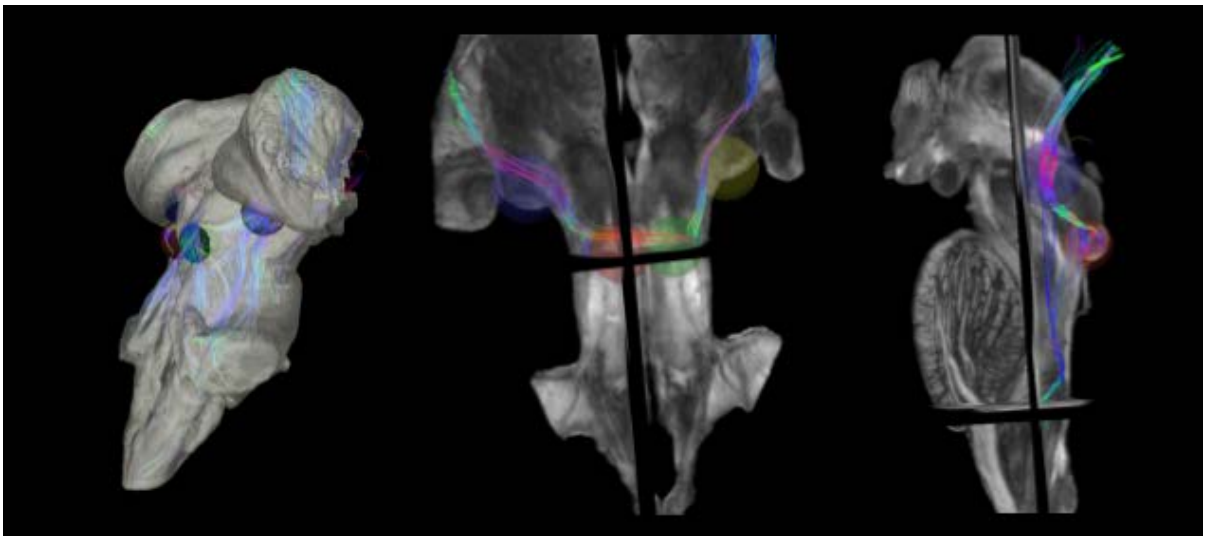
Support: NIH NRSA Fellowship F31 DC015695-02

NIH-NIBIB R01 EB020740

Title: Diffusion tractography of the subcortical auditory system in a postmortem human brain

Authors: *K. R. SITEK[1], E. CALABRESE[2], G. A. JOHNSON[3], S. S. GHOSH[1]
[1]MIT, Cambridge, MA; [3]Radiology, [2]Duke Univ., Durham, NC

Abstract: The auditory system connects the sensory epithelium of the inner ear to the auditory cortex and is comprised of multiple intermediate subcortical structures. Research in animal models has revealed numerous functional subdivisions of auditory structures and structural connections between these structures. However, investigating the anatomy of the auditory system in humans has been much more limited in scope due to technical challenges of imaging subcortical regions. Here, we use high-field diffusion-weighted MR microscopy of a postmortem human brain stem acquired at the Duke Center for In Vivo Microscopy to identify pathways of the human subcortical auditory system. Diffusion data of the specimen, which included the medulla, pons, midbrain, and thalamus, were acquired at 7 Tesla in 120 unique diffusion directions with b=4000 s/mm^2 and 200 μm isotropic resolution. White matter tractography was performed using model-free reconstruction based on diffusion orientation distribution functions at each voxel. By creating seed regions of interest, we could clearly identify white matter pathways and connections between subcortical auditory structures. These include the lateral lemniscus tract between the brainstem and the inferior colliculus, fibers of the cochlear nerve, and medial geniculate-IC connections. In addition to the high sensitivity and angular resolution of the diffusion data, the high spatial resolution allows us to segment subcortical auditory structures into subnuclei and trace their specific connections throughout the auditory pathway. To our knowledge, this is the highest quality tractography study of the human subcortical auditory system. We can apply findings from this study to lower resolution in vivo human imaging data to improve mapping of the auditory system in other research and clinical populations.



Disclosures: **K.R. Sitek:** None. **E. Calabrese:** None. **G.A. Johnson:** None. **S.S. Ghosh:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.04/GG24

Topic: D.06. Audition

Support: NIH DC007733 (to AGH)

1I01RX001095-01 U.S. Dept. of Veterans Affairs to AGH

Title: Noise induced hearing loss differentially reduces the dimerized pool of dopamine receptors and their interaction with N-ethylmaleimide-sensitive fusion (NSF-1) protein across auditory related brain regions

Authors: A. K. APAWU[1], A. DIXON[1], B. ADAMS[1], M. HALI[1], B. FYK-KOLODZIEJ[2], *A. HOLT[1]
[1]Anat. & Cell Biol, [2]Physiol., Wayne State Univ. Sch. of Med., Detroit, MI

Abstract: While hearing loss, tinnitus, and hyperacusis have been associated with temporal and spatial changes in spontaneous neuronal activity, the neurochemical bases for these changes remain elusive. Not only have we reported deafness related changes in dopamine (DA) receptor (Drd) gene expression, but have also shown that these changes are brain region and time dependent. Furthermore, our work has revealed that the function of DA receptors in the inferior colliculus (IC) is both activity driven and class specific (Drd1 or Drd2). In the present work, we further examined the role of Drds in hearing by investigating the effect of noise exposure on Drd

localization, protein levels, and activity dependent interaction with the trafficking protein NSF-1. Adult male Sprague Dawley rats (n=14) were divided into normal hearing and noise exposure groups (16 kHz,106 dB, SPL, 1 hour; and 10 kHz, 118 dB SPL, 1/3 octave band, 4 hours). Receptor localization was examined in cryosections (40 µm) obtained from the IC and auditory cortex (AC) using immunocytochemistry. Following protein isolation, Drd1 and Drd2 levels were compared across groups using Western blot (7 µg/lane). Co-immunoprecipitation with and without calcium was used to determine activity dependent DA receptor/NSF-1 interactions.
In the IC, Drd1 labeling filled the somata while Drd2 labeling was patchy, within somata and dendrites. In animals with a permanent threshold shift (PTS) after noise exposure, somatic labeling for Drd1 appeared to be increased compared to controls while labeling for Drd2 appeared to decrease within somata, but not processes. While both Drd1 and Drd2 appeared to form dimers (~100 kDa) in the IC and AC, Drd1 primarily exists in the dimerized state. One day following the PTS, dimerized Drd2 was significantly decreased in the AC. The Drd1-NSF-1 interaction in the IC was calcium dependent in animals with a temporary threshold shift (TTS), at a time when hearing thresholds had returned to normal. In the AC, TTS resulted in increased NSF-1/Drd interaction in the presence of calcium.
These results suggest DA receptors in the IC and AC are affected by noise induced permanent and temporary hearing loss even if hearing returns to normal. The spatial differences in the effects of noise on dopamine receptor production, dimerization, and trafficking suggest functional differences in dopaminergic mechanisms between auditory brain regions.

Disclosures: **A.K. Apawu:** None. **A. Dixon:** None. **B. Adams:** None. **M. Hali:** None. **B. Fyk-Kolodziej:** None. **A. Holt:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.05/GG25

Topic: D.06. Audition

Support: Howard Hughes Institute Collaborative Award

ERC Grant

US-Israel Bi-national Grant

Title: Genetic access to active neurons in the mouse auditory cortex

Authors: *G.-I. TASAKA[1], C. GUENTHNER[2], A. SHALEV[1], O. GILDAY[1], M. GROYSMAN[1], L. LUO[2], A. MIZRAHI[3]
[1]Dept. of Neurobiology, ELSC, The Hebrew Univ. of Jerusalem, Jerusalem, Israel; [2]Dept. of

Biol., Howard Hughes Med. Inst. - Stanford Univ., Stanford, CA; [3]Hebrew Univ. of Jerusalem, Jerusalem, Israel

Abstract: Cortical circuits are often functionally heterogeneous and their responses can be sparse. In the auditory system, it is difficult to access specific local circuits encoding stimuli such as natural sounds as these shape neuronal responses in complex ways; for example, due to their rich frequency content, temporal structure and contextual effects. Here, we used a new method, called TRAP (*Targeted Recombination in Active Populations*) to identify and access functionally active circuits in the mouse auditory cortex (A1), *in vivo*. TRAP is based on a Fos-CreER^{T2} driver line allowing genetic access of temporarily active neurons. We developed and tested a new reporter mouse strain (called TB), which allows the expression of a fluorescent protein and tTA after the excision of a loxP-flanked transcriptional stop signal. The new reporter enables labeling of neurons as well as induction of other genes when these are expressed under the TRE promoter. We calibrated the FosCreER^{T2}xTB system to A1. We demonstrate near zero non-specific labeling in A1 and show up to 3-fold increase in the number of trapped neurons following sound exposure to natural sounds (but not pure tones). Efficient recombination is also shown in other regions of the auditory pathway. In vivo two-photon targeted patch recording from L2/3 neurons shows that TRAPed neurons respond significantly stronger to the trapping stimulus as compared to non-TRAPed neighbors or as compared to control TRAPed neurons. Next, we used this system to investigate plasticity of postpartum mothers. Pup ultrasonic vocalizations (USV) induced higher numbers of recombined neurons in mothers' A1 compared with naïve virgins. USV-TRAPed neurons in mothers were found to have a unique spiking signature suggesting that a newly recruited subpopulation of cortical neurons contribute to efficient encoding of USV. Our method will now be used as a substrate to elucidate the unique neural signature of parental plasticity across the auditory system.

Disclosures: G. Tasaka: None. **C. Guenthner:** None. **A. Shalev:** None. **O. Gilday:** None. **M. Groysman:** None. **L. Luo:** None. **A. Mizrahi:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.06/GG26

Topic: D.06. Audition

Support: National Science Foundation Grant 1208131

Title: Mechanisms of changes in synaptic depression in response to different levels of activity at endbulbs of Held

Authors: *X. ZHUANG[1], W. SUN[2], M. A. XU-FRIEDMAN[1]
[1]SUNY UNIVERSITY AT BUFFALO, Buffalo, NY; [2]Dept. of Communicative Disorders and Sci., SUNY Univ. at Buffalo, Buffalo, NY

Abstract: Abnormal levels of auditory activity trigger an adaptation mechanism that changes the properties of endbulbs of Held, which are synapses formed by auditory nerve fibers onto bushy cells in the anteroventral cochlear nucleus. After exposure to an augmented acoustic environment, endbulbs of young mice reduce their synaptic depression, while after a week of ear occlusion, they show greater synaptic depression. It is important to understand the mechanisms underlying these changes, because abnormal auditory activity is associated with hearing problems including tinnitus and processing disorders. The most likely mechanism involves changes in presynaptic calcium, either changes in calcium influx, or in the efficacy of calcium driving neurotransmitter release. We tested these possibilities using voltage-clamp recordings of bushy cells in mouse brain slices. Bath-application of the slow chelator EGTA-AM blocked EPSCs more effectively in augmented endbulbs than in control endbulbs. This is consistent with decreased calcium influx or reduced coupling efficacy between calcium channels and the release apparatus, which could account for decreased synaptic depression. The fast chelator BAPTA-AM was less effective at blocking EPSCs in occluded endbulbs, which is consistent with tighter coupling or an increase in calcium influx and could account for increased synaptic depression. We also tested changes in coupling efficacy by quantifying the cooperativity following exposure to different sound environments and found no change. We plan to further assess calcium influx using calcium imaging. Another question is whether activity adjusts endbulbs only during development. We found that mature endbulbs (>postnatal 50 days) showed activity-dependent changes in synaptic depression similar to young endbulbs. This suggested there is no critical period for this phenomenon and endbulbs adjust their properties as auditory nerve activity changes throughout life. By understanding the mechanism of changes in synaptic depression, it may provide new ideas for treating disorders such as tinnitus and conductive hearing loss.

Disclosures: X. Zhuang: None. **W. Sun:** None. **M.A. Xu-Friedman:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.07/DP08/GG27 (Dynamic Poster)

Topic: D.06. Audition

Support: IARPA MiCRONS

Title: Integrating in-vivo imaging and neuronal barcoding to link neural coding and network structure in mouse cortex

Authors: *A. VAUGHAN, C. J. STONEKING, A. M. ZADOR
Cold Spring Harbor Lab., Cold Spring Harbor, NY

Abstract: In recent years, there has been considerable progress in understanding the mesoscale connectome of the mouse brain using neuronal barcoding (Kebschull et al., 2016). Parallel efforts have sought to develop high-throughput recordings of neural activity in order to understand the information geometry of sensory processing, decision-making, and motor control. To date, however, high throughput methods for each field - whole-brain connectomics and neural recording - have been fundamentally incompatible, leading to a piecemeal understanding of neural circuit form and function. Here, we describe a method that integrates GCaMP imaging with neuronal barcoding that enables the simultaneous resolution of both the activity and mesoscale connectivity of thousands of neurons simultaneously. Using GCaMP recordings of neurons in primary auditory cortex, we delineate the functional response properties of thousands of neurons using a suite of auditory stimuli. We then use neuronal barcoding to map the projections of these neurons, using a combination of mapSEQ (Kebschull et al., 2016) with in-situ sequencing (Lee et al., 2014; Chen et al., in prep). Linking these two datasets with high-resolution registration of in-vivo and ex-vivo data, we are able to identify and integrate the functional response properties and projection anatomy of thousands of neurons simultaneously. Using this approach, we generate an integrated view of diversity in neuronal activity as well as morphology, providing a unique holistic view into cortical structure and function.

Disclosures: A. Vaughan: None. **C.J. Stoneking:** None. **A.M. Zador:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.08/GG28

Topic: D.06. Audition

Support: NIH Grant U01NS090569

Title: Reliable and persistent population encoding of sounds across layers in the auditory cortex of awake mice

Authors: *D. E. WINKOWSKI[1], *D. E. WINKOWSKI[1], Z. BOWEN[2], T. L. RIBEIRO[3], D. PLENZ[4], P. KANOLD[1]
[2]Biol., [1]Univ. of Maryland, College Park, MD; [3]Section on Critical Brain Dynamics LSN/NIMH, NIH, Bethesda, MD; [4]Sect Critical Brain Dynamics, Natl. Inst. of Mental Health, NIH, Bethesda, MD

Abstract: The ability to detect and encode sensory stimuli hinges on the encoding capability of neuronal populations. Individual neurons in primary auditory cortex (A1) can show selectivity to stimulus features (i.e., sound frequency). However, the representation of sound information in populations of A1 neurons and the nature of populations activated by sound stimuli is unresolved. The theory of critical dynamics describes complex systems that operate at or near a critical point, i.e., balanced between order and disorder, where systems gain advantages in information processing and their dynamics become scale-invariant. This theory has been gaining traction in neuroscience since the identification of neuronal avalanches, intermittent spatiotemporal activity patterns with scale-invariant properties, e.g. their size and duration, are power law distributed.
Here, we tested whether sound processing in A1 neuronal populations of Layer 2/3 (L2/3) and Layer 4 (L4) is in line with expectations for critical dynamics. We characterized spontaneous as well as sound evoked activity in L2/3 and L4 in A1 of awake mice using in vivo 2-photon calcium imaging of neuronal populations. We explore the relationship between properties of sensory stimuli, single neuron responses, and population responses with respect to avalanches and compare our findings across cortical laminae. We investigate the spectral tuning properties of neurons participating in avalanches in order to probe the relationship between criticality and sensory coding. On the population level, we found that avalanche statistics varied with stimulus frequency and intensity of the presented sound. In L2/3, we found that avalanche patterns reliably represented sounds independent of level and contained stimulus information well after sound offset, revealing long-range spatiotemporal activity correlations not evident at single-cell or cell-pair level. Ongoing work is investigating whether this persistent encoding of stimulus information is inherited from L4 populations or whether this is an emergent feature of population activity in L2/3.
Our investigation will provide insight into how neural networks containing differing populations of neurons with varying firing rates stably encode information about sensory stimuli in the context of critical dynamics.

Disclosures: D.E. Winkowski: None. **Z. Bowen:** None. **T.L. Ribeiro:** None. **D. Plenz:** None. **P. Kanold:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.09/GG29

Topic: D.06. Audition

Support: the National Center of Competence in Research (NCCR) ‘‘SYNAPSY—The Synaptic Bases of Mental Diseases’’ financed by the Swiss National Science Foundation (Grant no.51AU40_125759)

Title: Feedforward and feedback mechanisms governing auditory sensory gating in awake mice

Authors: *A. KHANI[1], F. LANZ[1], K. SCHALLER[2], C. M. MICHEL[1], C. QUAIRIAUX[1]
[1]Functional Brain Mapping Laboratory, Dept. of Basic Neurosciences, [2]Geneva Univ. Hospitals, Fac. of Med., Univ. of Geneva, Geneva, Switzerland

Abstract: In association with impaired performance in attention related cognitive tasks, EEG studies of schizophrenic patients repeatedly showed alterations in processing of auditory stimulation and in particular in auditory sensory gating and mismatch negativity. Sensory gating refers to the fundamental property of the brain that down-regulates neural response to a stimulus that is repeated after a short interval. Sensory gating is thought to reflect an inhibitory processing component of attention; however despite the long history of characterization of this phenomenon, the neural mechanisms are not clearly understood. One hypothesis is that sensory gating is governed by top-down influences from the prefrontal and cingulate areas on the bottom-up sensory processes.
We addressed neural mechanisms of sensory gating in head-fixed awake mice using either surface EEG or simultaneous multi-site intracerebral recordings from the inferior colliculus centralsis (ICc), auditory thalamus (medial geniculate nucleus; MGN), primary auditory cortex (Au1) and anterior cingulate cortex (ACC). We used paired tone paradigm with a wide frequency range (white noise) and a variable inter-stimulus interval (ISI). Analysis of global field power from surface EEG recordings revealed sensory gating up to 2s ISI and pointed to the activation of a large-scale auditory network propagating from the brainstem to the frontal cortices. Our intra-cortical recordings showed a robust attenuation of the LFP amplitude and spike rates of a subset of single neurons immediately after the second tone. The early sensory gating was also reflected in reduced gamma power in the ICc, MGN and Au1 suggesting a feedforward mechanism for attenuation of response from the ICc through MGN to Au1. On the other hand, a delayed increase in beta power in thalamus and delayed firing (~150 ms) of a subset of single neurons in the MGN and Au1 auditory cortex following the first tone suggest a top down mechanism involved in the regulation of sensory gating. This was further supported by a robust enhancement of the cross frequency phase-amplitude coupling between phases of low frequency (~15-20 Hz) LFP in the ACC and amplitude of gamma (~40-60 Hz) power in the ICc. Preliminary results of optogenetic manipulations suggest a causal role for top-down regulation of sensory gating. Taken together, our results suggest a differential contribution of both feedforward and feedback mechanisms in regulating sensory gating.

Disclosures: A. Khani: None. **F. Lanz:** None. **K. Schaller:** None. **C.M. Michel:** None. **C. Quairiaux:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.10/GG30

Topic: D.06. Audition

Support: CIHR

NSERC

Title: Effect of ACh and general DREADD inhibition in tegmental areas on sensorimotor gating

Authors: *N. FULCHER[1], E. C. AZZOPARDI[2], C. DE OLIVEIRA[2], S. SCHMID[2]
[1]Neurosci., The Univ. of Western Ontario, London, ON, Canada; [2]Univ. of Western Ontario, London, ON, Canada

Abstract: Humans have intricate brains that deal with an abundance of stimuli emerging from the environment at all times. To prevent overwhelming our brains with sensory information, sensorimotor gating allows for the filtering of redundant information. Sensorimotor gating deficits are seen in an array of neurological disorders such as autism spectrum disorder, schizophrenia, and others. An established way to quantify sensorimotor gating is through prepulse inhibition (PPI) of the acoustic startle response (ASR). Here, we look to further understand mechanisms underlying PPI and its disruptions, to better comprehend how they are involved in affected neurological diseases.
The midbrain has been established as an integral brain region mediating PPI of the ASR. The acoustic pathway involved in PPI consists of: cochlear root neurons, the cochlear nuclei, the inferior (IC) and superior colliculi (SC), the laterodorsal (LDT) and pedunculopontine tegmental nuclei (PPT). The LDT and PPT neurons project to the startle mediating giant neurons of the PnC, which is a crucial part of the complex startle pathway. Lesions to the LDT severely attenuate PPI in rats, while lesions within the PPT disrupt PPI.
Designer Receptor Exclusively Activated by Designer Drugs (DREADDs) is an innovative chemogenetic technique that involves the delivery of viral constructs intracranially and allows drug injections to transiently activate/inhibit targeted classes of neurons. These viral constructs carry a specified DREADD receptor, promoter element, and fluorescent reporter that are introduced into the neuronal membrane. Here, we intracranially delivered bilaterally either the Cre-dependent inhibitory DREADD, or a control vector, into either the PPT or LDT of Cre-ChAT male and female adult Long-Evans rats. Subjects recovered for 3 weeks and received an i.p. injection of either the DREADD ligand clozapine N-oxide (CNO) or vehicle. Immunohistochemistry verified the effectiveness of transfection and quantified co-localization of the DREADD receptors with ACh markers.
Our findings suggest that DREADD-induced inhibition of ACh within the PPT or LDT does not

mediate PPI. Since both structures are composed of a heterogeneous collection of glutamate, ACh, and gamma-aminobutyric acid (GABA) neurons, future work will involve the inhibition of all three cell types in both regions to advance understanding of the potentially crucial role of the tegmental areas in sensorimotor gating. This work will help to develop better-suited drugs to enhance prepulse inhibition patients with deficits in sensorimotor gating.

Disclosures: N. Fulcher: None. **E.C. Azzopardi:** None. **C. De Oliveira:** None. **S. Schmid:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.11/GG31

Topic: D.06. Audition

Support: NIH Grant DC004682

NIH Grant DC015239

Title: Behavioral state modifies excitation and inhibition in auditory cortical neurons

Authors: *N. EDWARDS[1], J. S. ISAACSON[2]
[1]Neurosciences, Uinversity of California, San Diego, LA Jolla, CA; [2]Neurosciences, UCSD, La Jolla, CA

Abstract: Factors such as arousal or attention are likely to modulate the processing of sensory information in cortical circuits. Indeed, recent studies indicate that changes in brain state are coupled to changes in the excitability of pyramidal cells in sensory cortices. While previous studies have relied on current clamp recordings to monitor changes in membrane potential, how changes in brain state alter excitatory and inhibitory input to cortical pyramidal cells is unclear. Here, we use whole-cell voltage clamp recordings from layer 2/3 pyramidal cells in the primary auditory cortex (A1) of awake mice to determine the effect of behavioral state on excitation and inhibition. We monitored pupil diameter as a measure of arousal while recording spontaneous and sound-evoked excitatory and inhibitory synaptic inputs. We find that increases in pupil diameter (increases in arousal) are coupled to decreases in both inhibition and excitation. However, brain state-dependent changes in inhibitory and excitatory input appear asymmetric. Ongoing experiments should provide insight into the synaptic and circuit mechanisms underlying behaviorally-driven changes in pyramidal cell excitability.

Disclosures: N. Edwards: None. **J.S. Isaacson:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.12/GG32

Topic: D.06. Audition

Support: NSF GRFP 2015215385

NIH DC004682

NIH DC015239

Title: Characterizing the organization and function of auditory cortex projection neurons

Authors: *P.-A. LIN, J. S. ISAACSON
Neurosciences, UCSD, San Diego, CA

Abstract: Sensory information processed in the primary auditory cortex (A1) is directed to a variety of spatially and functionally distinct targets including contralateral A1, inferior colliculus (IC), striatum, and thalamus. While these target regions play distinct roles in behavior and cognition, the properties of A1 neurons projecting to these structures and the information they convey are not well understood. We set out to determine whether individual A1 projection neurons serve as generalists that send redundant auditory information to several downstream regions, or as specialists that transmit particular aspects of auditory information to a single downstream region. To address this question, we used the retrograde tracer cholera toxin B (CTB) conjugated to different fluorescent dyes to simultaneously label and visualize multiple target-specific subpopulations of A1 projection neurons in the adult mouse brain. We find that neurons projecting to the inferior colliculus very rarely project to other targets. Furthermore, we find only a small subset of pyramidal cells targeting contralateral A1 that also project to the striatum. Taken together, our results indicate that the vast majority of A1 projection neurons act as specialists projecting to single downstream targets. We are currently using vivo calcium imaging in awake mice to determine the sound-evoked response properties of pyramidal cells that project to different targets.

Disclosures: P. Lin: None. **J.S. Isaacson:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.13/GG33

Topic: D.06. Audition

Support: NIH Grant EY022720

NIH Grant DC009607

NIH Grant GM056481

Title: Sublaminar subdivision of mouse auditory cortex layer 2/3 based on functional connections

Authors: *X. MENG[1], J. P. Y. KAO[2], P. O. KANOLD[1]
[1]Biol., Univ. of Maryland, College Park, MD; [2]Ctr. for Biomed. Engin. and Technology, and Dept. of Physiol., Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: The cerebral cortex is subdivided into 6 layers based on morphological features. In particular, the supragranular layers 2/3 (L2/3) are of interest because they reflect the first hierarchical cortical processing stage after sensory input is received in layer 4 (L4). In the auditory cortex (A1) sound frequency information is represented tonotopically in L4 but the frequency organization of supragranular L2/3 is more heterogeneous suggesting an interlaminar transformation of sound frequency representation. L2/3 contains morphological and genetically diverse population of neurons suggesting that the functional heterogeneity in L2/3 could be the result of cellular or circuit diversity.
We here investigated in mouse auditory cortex if L2/3 contains discrete subpopulations of cells with specific functional microcircuits. We use in vitro slice recordings coupled with laser scanning photostimulation (LSPS) to reveal the intracortical excitatory and inhibitory microcircuits impinging on L2/3 neurons. We then use hierarchical clustering on the excitatory and inhibitory laminar connection patterns. We find that multiple distinct classes of L2/3 neurons exist. The classes of L2/3 neurons are distinguished by their laminar patterns of inputs from within A1, their location within L2/3, and the amount of integration across the tonotopic axis. These results suggest that similar to higher mammals, rodent L2/3 is not a homogenous layer but that several parallel microcircuits exist.

Disclosures: X. Meng: None. **J.P.Y. Kao:** None. **P.O. Kanold:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.14/HH1

Topic: D.06. Audition

Support: European Research Council Grant no. 340063 (project RATLAND)

Human Frontiers Science Program Grant RGP0044/2015

Title: Optogenetic manipulation of inhibitory populations in auditory cortex during stimulus-specific adaptation

Authors: *T. S. YARDEN, A. MIZRAHI, I. NELKEN
Hebrew Univ., Jerusalem, Israel

Abstract: Neurons in primary auditory cortex exhibit stimulus-specific adaptation (SSA), the decrease in responses to a common (standard) stimulus that does not generalize fully to another, rare stimulus (deviant). We investigate the role of inhibition in shaping SSA using optogenetic manipulation of the activity of specific inhibitory populations in auditory cortex. We used a viral vector for conditional expression of the Arch opsin in transgenic mice, and performed *in vivo* two-photon targeted loose-patch recordings, targeting both pyramidal neurons and opsin-expressing inhibitory neurons. We show that the optogenetic effects on the opsin-expressing neurons were sometimes non-trivial, e.g. increasing the sensory response rather than decreasing it, or decreasing it more strongly at lower light intensities. Optogenetic effects depended on the protocol of sensory stimulation and on the intensity of light stimulation, and were different for spontaneous activity and for different phases of the sensory-evoked activity. In addition, we relate the manipulation of inhibitory neuron activity to the responses recorded in pyramidal neurons, showing the contribution of inhibition to the excitatory cortical activity during SSA

Disclosures: T.S. Yarden: None. **A. Mizrahi:** None. **I. Nelken:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.15/HH2

Topic: D.06. Audition

Title: GABAB receptors in cortical input layers mediate spike-timing dependent LTD of inhibition and regulate critical period plasticity

Authors: *E. D. VICKERS[1], C. CLARK[2], A. FRATZL[2], R. SCHNEGGENBURGER[2]
[2]Brain Mind Inst., [1]EPFL, Lausanne, Switzerland

Abstract: The tonotopic representation of pure tones in the input layers of primary auditory cortex depends on sound experience during a "critical period" (CP) of brain development. Parvalbumin (PV) - expressing cortical interneurons have been strongly implicated in CP plasticity. We have previously shown that PV interneuron output synapses onto input layer principal cells (PC) in auditory cortex exhibit bi-directional spike-timing dependent (STD) plasticity (SfN 2016 abstract: Vickers and Schneggenburger; 401.13/G11). Interestingly, long-term depression of inhibition (iLTD), observed with pre- (PV interneuron) before postsynaptic (PC) spike-timing orders, depended on $GABA_B$ receptor signalling, was converted into iLTP both by further brain development (P25 - P28), and by sound exposure during the CP (P11 - P14).
To investigate possible links between iLTD and CP plasticity, we knocked-out $GABA_B$ receptor expression in PCs of the input layers. For this purpose, we crossed $Scnn1a^{Cre}$ mice with floxed $GABA_BR1$ mice (Haller et al, 2004) and tdTomato reporter mice. This allowed us to perform paired whole-cell recordings from tdTomato+ PCs and their connected PV-interneurons. In PCs lacking $GABA_BR1$, we found that iLTD was eliminated at P18-20, revealing a specific role for postsynaptic $GABA_B$-receptors in the induction of iLTD. In addition, the baseline IPSCs amplitudes in PV-IN to PC pairs were significantly smaller in $GABA_BR1$ cKO mice.
We next wished to study the role of $GABA_B$ receptors in PCs of the input layers in CP band expansion. For this, we crossed $Scnn1a^{Cre}$ x $GABAB_R1^{lox/lox}$ mice with cfos-tTA x tetO-tdTomato mice. This enabled us to use 2-photon imaging of tdTomato in thalamocortical brain slices to quantify the band of neurons activated by 30 kHz tones. We found a band of cFos-labelled neurons in naive $GABAB_R1$ cKO mice. However, in $GABAB_R1$ cKO mice that had been exposed to CP sound conditioning for 6 days (P12-18), the band of cFos-labeled neurons was reduced in size and contained a lower density of labeled cells. Thus, we speculate that, following CP conditioning in the absence of postsynaptic $GABA_B$Rs, there is a misregulated strengthening of inhibition, maybe caused by the absence of iLTD, which then leads to suppressed tone-activation of PCs.

Disclosures: E.D. Vickers: None. **C. Clark:** None. **A. Fratzl:** None. **R. Schneggenburger:** None.

Poster

399. Auditory Processing: Circuits, Synapses, and Neurotransmitters II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 399.16/HH3

Topic: D.06. Audition

Title: Cortical circuits of GABAergic callosal projections

Authors: *C. ROCK, H. ZURITA, S. LEBBY, C. J. WILSON, A. J. APICELLA
Biol., Univ. of Texas at San Antonio, San Antonio, TX

Abstract: A deep-rooted principle of cortical circuit organization is that excitatory neurotransmission is involved in both local and long-range processing, whereas inhibition is exclusively local. In favor of this, anatomical studies have shown that the majority of axons projecting via the corpus callosum are glutamatergic. However, a small but significant proportion of callosal axons are also immunoreactive for glutamic acid decarboxylase, an enzyme required for gamma-aminobutyric acid (GABA) synthesis and a specific marker for GABAergic neurons. In this study we tested the hypothesis that corticocortical parvalbumin-expressing (CC-Parv) neurons connect the two hemispheres of multiple cortical areas, project through the corpus callosum, and are a functional part of the local cortical circuit. Our investigation of this hypothesis takes advantage of viral tracing and optogenetics to determine the anatomical and electrophysiological properties of CC-Parv neurons of the mouse auditory, visual, and motor cortices. We found a direct inhibitory pathway made up of parvalbumin-expressing (Parv) neurons which connects corresponding cortical areas. Like other Parv cortical neurons, these neurons provide local inhibition onto nearby pyramidal neurons and receive thalamocortical input. Comparison of CC-Parv neurons to putative non-CC-Parv neurons shows a dissimilarity in both their morphological characteristics as well as their intrinsic properties. These differences in the intrinsic properties, such as instantaneous firing frequency, of CC-Parv neurons and putative non-CC-Parv neurons may reflect differential involvement in slow and fast gamma-band cortical oscillations, respectively.

Disclosures: C. Rock: None. **H. Zurita:** None. **S. Lebby:** None. **C.J. Wilson:** None. **A.J. Apicella:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.01/HH4

Topic: D.06. Audition

Support: NIH DC012833

NIH MH097742

ALSAC

Title: Restoring neuronal plasticity in the auditory cortex in adult mice by reducing thalamic adenosine signaling

Authors: *J. A. BLUNDON, N. ROY, B. TEUBNER, J. YU, S. HAN, S. ZAKHARENKO
St Jude Children's Res. Hosp, Memphis, TN

Abstract: Circuits in the auditory cortex are highly susceptible to acoustic influences during an early postnatal critical period. The auditory cortex selectively expands neural representations of enriched acoustic stimuli, a process important for human language acquisition. Adults lack this plasticity. Here we show in the murine auditory cortex that juvenile plasticity can be reestablished in adulthood if acoustic stimuli are paired with disruption of ecto-5'-nucleotidase-dependent adenosine production or A1 adenosine receptor signaling in the auditory thalamus. We used repeated two-photon imaging of Layer III/IV neuronal responses to sound stimuli in auditory cortex of awake mice to track neuronal best frequency during continuous sound exposure training. Neurons in adult mice with diminished or blocked thalamic adenosine signaling shift their best frequency towards the exposure frequency after only 24 hours of training. These mice also show long-term improvement of tone-discrimination abilities. We conclude that, in adult mice, disrupting adenosine signaling in the thalamus rejuvenates plasticity in the auditory cortex and improves auditory perception.

Disclosures: J.A. Blundon: None. **N. Roy:** None. **B. Teubner:** None. **J. Yu:** None. **S. Han:** None. **S. Zakharenko:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.02/HH5

Topic: D.06. Audition

Support: NIH Grant DC012833

NIH Grant MH097742

ALSAC

Title: Restoring auditory cortical map plasticity in adult mice by disrupting thalamic adenosine signaling

Authors: *N. ROY, J. A. BLUNDON, B. TEUBNER, J. YU, T.-Y. EOM, S. HAN, S. ZAKHARENKO
Developmental Neurobio., St. Jude Children's Res. Hosp., Memphis, TN

Abstract: Circuits in the auditory cortex are shaped by acoustic influences during an early postnatal critical period. During this period, neural representations of frequently-occurring acoustic stimuli expand, a process which may be important for human language acquisition. Passive exposure to acoustic stimuli does not lead to cortical plasticity in adults. Here we show in mouse auditory cortex that the juvenile form of plasticity can be reestablished in adulthood if acoustic stimuli are paired with disruption of ecto-5'-nucleotidase-dependent adenosine production or A_1 adenosine receptor signaling in the auditory thalamus. This plasticity occurs at the level of cortical maps and individual neurons in the auditory cortex of awake adult mice and is associated with long-term improvement of tone-discrimination abilities. We conclude that, in adult mice, disrupting adenosine signaling in the thalamus rejuvenates plasticity in the auditory cortex and improves auditory perception.

Disclosures: N. Roy: None. **J.A. Blundon:** None. **B. Teubner:** None. **J. Yu:** None. **T. Eom:** None. **S. Han:** None. **S. Zakharenko:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.03/HH6

Topic: D.06. Audition

Support: Fondation Fyssen Postdoctoral Fellowship

Title: Synaptic and spiking responses to infant vocalizations in mouse paraventricular hypothalamus *In vivo*

Authors: *S. VALTCHEVA, R. C. FROEMKE
Skirball Inst., NYU Sch. of Med., New York, NY

Abstract: Experience-driven changes in neural circuits are believed to have important consequences for information processing and sensory perception. A fundamental question is how neural plasticity occurs under natural conditions, to promote the recognition of stimuli with behavioral significance. Motherhood is a dramatic natural experience (Dulac et al., 2014), but little is known about the underlying changes in neural circuits for parenting behavior. The neuropeptide oxytocin is synthesized by hypothalamic areas including the paraventricular nucleus (PVN), and is important for maternal behaviors including parturition, lactation, and parent-child bonding, perhaps in part by increasing salience of social information (Insel & Young, 2001). Indeed, many women experience oxytocin release and milk ejection simply in response to infant cries (McNeilly et al., 1983). Recently, we showed that oxytocin can enable maternal behavior by balancing synaptic inhibition with excitation in the auditory cortex (Marlin et al., 2015; Mitre et al., 2016). Therefore, cortical modulation and plasticity produced by

oxytocin signaling might enable long-lasting enhancements of social interactions and maternal care. However, beyond the necessity for lactation, mechanisms of oxytocin release in the brain leading to naturally-induced forms of plasticity are not well understood. Here, we aimed at identifying how social stimuli activate PVN oxytocin neurons in vivo, and if these responses were gated or changed after maternal experience. We performed in vivo cell-attached and whole-cell recordings from PVN neurons in awake head-fixed mice. We examined PVN neuronal responses to ultrasonic vocalizations (pup isolation calls) and pure tones. PVN neurons did not display frequency selectivity in response to pure tones but they showed reliable responses to pup calls. However, in virgin females these responses remained predominantly subthreshold. Our results suggest that PVN neurons can receive information about acoustic input, reflecting projections from central auditory areas. In absence of maternal experience, however, these responses are unreliable and less coherent, keeping the synaptic responses subthreshold and preventing spurious action potential generation and subsequent oxytocin release in naïve animals.

Disclosures: S. Valtcheva: None. **R.C. Froemke:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.04/HH7

Topic: D.06. Audition

Support: National Science Foundation GRFP

Title: Perceptual categorization of pup vocalizations in the auditory cortex of maternal mice

Authors: *J. SCHIAVO[1], R. C. FROEMKE[2]
[1]New York Univ., New York, NY; [2]Otolaryngology, NYU Med., New York, NY

Abstract: Learning requires generalization from specific exemplars to enable animals to respond reliably to different stimuli with the same behavioral significance. Perceptual categorization of vocalizations facilitates communication by grouping stimuli into discrete categories despite variability in acoustic features (such as pitch or tempo) between individuals. Little is known about the mechanisms underlying learned category formation at the single-cell and population level, as well as the link between categorical representations and generalization from learned categories. Here we take advantage of a natural behavior in adult mice: retrieval of nest-isolated pups based on distress calls (Ehret Nature 1987, Liu & Schreiner PLoS Biol 2007, Cohen et al. Neuron 2011, Marlin et al. Nature 2015). Naive virgin females initially do not behaviorally respond to these calls, while mothers and virgins co-housed with dams and litters can rapidly respond to distress calls by bringing the pup back to the nest. These ultrasonic vocalizations

(USVs) have distinct spectro-temporal features, occurring in bouts of 3-8 Hz with spectral peaks at 40-80 kHz. USV sequences are variable between and within individuals, however the behavioral significance is the same: to signal the mother. This behavior requires females to perceptually categorize USVs, and serves as a model to examine the neural mechanisms underlying category formation in the auditory cortex. Here we used behavioral measurement and 2-photon imaging to monitor changes in neural and behavioral categories. We assessed the categorical boundary for pup calls in the frequency and temporal domain by morphing pre-recorded USVs from P1-8 pups. First, we used these morphs to behaviorally assess response boundaries in maternal mice using a Y-maze. Mothers reliably approached speakers playing pre-recorded USVs, and approached speakers playing USVs modulated at the ethological rate when compared to time-warped USVs. Using 2-photon imaging in awake mice, we assessed neural response boundaries for pup calls in maternal and non-maternal females. Excitatory neurons in maternal auditory cortex responded invariantly to time-warped and pitch-shifted calls within the natural range, while excitatory neurons in naïve females did not respond invariantly to morphed calls in either domain. The behavioral and neural data thus far is indicative of distinct cortical differences in the processing of USVs between maternal and naïve females, providing an opportunity to further study how the brain shifts from a non-maternal to maternal state to enable parental behaviors.

Disclosures: **J. Schiavo:** None. **R.C. Froemke:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.05/HH8

Topic: D.06. Audition

Support: NIH R01DC014656

NIH F32DC014376

Title: Cortical mechanisms of perceptual learning in juveniles

Authors: *M. L. CARAS, D. H. SANES
Ctr. for Neural Sci., New York Univ., New York, NY

Abstract: From riding a bike to learning a language, children are considered to be better skill learners than adults. One possible explanation for this superiority is that sensory systems display heightened plasticity during development, and thus may be more responsive to training. Developmental studies have documented differences between juvenile and adult learning, and have identified critical periods during which neural properties are particularly sensitive to

environmental experience. However, the neural plasticity that accompanies juvenile learning remains unexplored. To address this issue, we recorded telemetrically from left auditory cortex (ACx) of juvenile gerbils as they trained and improved on an amplitude modulation detection task. Neurometric and psychometric sensitivity were simultaneously tracked across days. As performance improved, juvenile ACx units displayed significant within-animal correlations between neural and behavioral thresholds. These data will be compared to similar recordings from mature adult animals to evaluate whether or not plasticity mechanisms differ.

Disclosures: M.L. Caras: None. **D.H. Sanes:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.06/HH9

Topic: D.06. Audition

Support: APA RD-2015-5A

APA RD-2015-5B

Title: Network-level modifications induced by sound-reward or -punishment associations in mice auditory cortex

Authors: *J.-F. LEGER[1], X. LIU[1], A. LOURDIANE[1], C. VENTALON[1], L. BOURDIEU[1], Y. BOUBENEC[2], S. A. SHAMMA[3,4]
[1]CNRS- Ecole Normale Superieure, Paris, France; [2]Lab. des Systèmes Perceptifs, Ecole Normale Superieure, Paris, France; [3]Univ. of Maryland, Washington, MD; [4]Lab. des Systèmes Perceptifs, Ecole Normale Supérieure, Paris, France

Abstract: Behavioral training on a task associating a sound with a reward or a punishment leads to extensive changes in the tonotopic map of the rodent auditory cortex (e.g. Rutkowsky & Weinberger, PNAS 2005, Shepard et al., eNeuro 2016). However, the corresponding changes in specific populations of neurons remain poorly described due to the difficulty of tracking the activity of defined neurons and populations throughout the learning process. We have used chronic two-photon calcium imaging with GCamp6f expression in layers 2/3 of the mouse primary auditory cortex to identify these changes after the animals learned a water-rewarded or electric shock-punishment operant sound recognition task. The tasks consisted in placing the animal in a shuttle box, presenting a target sound, and requiring the animal to switch its position in the box to either receive a water reward or escape an electric shock. This design was chosen to allow for direct comparison between the changes induced by positive and negative sound-meaning valence, without modification of the motor response in the tasks. Animals readily

learned the tasks and reached expert levels within 4 days (sound / electric shock association) to 10 days (sound / water reward association), succeeding in learning the two tasks sequentially. Calcium imaging performed before and after each learning period revealed profound modifications of individual cell responses, in particular with an average increase of the responses to the target sound, as well as the modifications induced at the cortical auditory network level.

Disclosures: **J. Leger:** None. **X. Liu:** None. **A. Lourdiane:** None. **C. Ventalon:** None. **L. Bourdieu:** None. **Y. Boubenec:** None. **S.A. Shamma:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.07/HH10

Topic: D.06. Audition

Support: NIH Grant No. R01 DC005779

Title: Categorical memory representation in ferret auditory and frontal cortices

Authors: *P. YIN[1], J. B. FRITZ[1,2], S. A. SHAMMA[1,2,3]
[1]Inst. For Syst. Research, Univ. Maryland, College Park, MD; [2]Electrical and Computer Engin. Department, Univ. of Maryland, College park, MD; [3]Sensory Perception Laboratory, Ecole Normale Superieure, Paris, France

Abstract: Categorization can arise by grouping of sensory stimuli sharing perceptual similarity or by training in which sensory stimuli are assigned to behaviorally relevant classes associated with learned responses. Neuronal category representations have been previously identified in vision in prefrontal, inferotemporal and parietal cortices, but a clear demonstration in audition has been elusive. In previous work, we showed that ferrets could be trained to classify auditory stimuli distributed along continuous feature dimensions (e.g. frequency or amplitude modulation) into classes based on behavioral meaning (Go or No-Go). Here we explore how these categories become represented in different fields of ferret auditory cortex (A1 and secondary areas in dPEG) and frontal cortex (FC), when animals were passively listening to sounds or actively engaged in task. To investigate the underlying mechanisms of category representation, we distinguish between “category-selective coding” and “sensory-selective coding”. Specifically, we expect stimuli from the same category to evoke shared response patterns across the neural population despite potentially large sensory differences between them, e.g., in perceived pitch or modulation. Two metrics based on the Euclidean distance of the population response between stimulus pairs were computed to evaluate these categorical effects: (a) categorical index (***CI***) - defined as the difference between the mean distances of the pairs the stimuli from different categories and of the pairs the stimuli from same category; and (b) the proportion of variance

explained by the categories (η^2). This analysis revealed that categorical information (Go *versus* No-Go) was observed in all three regions during task performance. Furthermore, the dynamics of this representation reveals that the categorical information emerges earliest in FC neurons, followed in dPEG, and then in primary A1. These results are consistent with the findings from visual system and provide insight into how the primary and secondary auditory cortices are differentially involved in classifying acoustic inputs during active auditory memory retrieval.

Disclosures: P. Yin: None. **J.B. Fritz:** None. **S.A. Shamma:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.08/HH11

Topic: D.06. Audition

Support: LIN special project

DFG projects He1721/10-1

DFG projects He1721/10-2

Title: Representations of tones in core fields of auditory cortex depend on their associated, upcoming behavioral outputs during the performance of auditory tasks

Authors: *Y. HUANG, M. BROSCH
Leibniz Inst. For Neurobio., Magdeburg, Germany

Abstract: Representations of sensory stimuli in sensory cortex depend on a variety of factors that are crucial for converting the stimuli into appropriate behavioral outputs during goal-directed tasks. However, an understanding of the effects of behavioral outputs themselves on sensory representations has been elusive. Here, by conducting a study in two nonhuman primates in which they performed two auditory tasks, we show that tone representations in core fields of auditory cortex depend on the upcoming behavioral outputs associated with the tones. The tasks were performed on the same set of stimuli and had a symmetrically rewarded go/no-go structure. The tasks and the stimuli were designed in such a way that the same tones could be associated with different behavioral outputs, i.e., making a bar release (*go response*) or withholding a bar release (*no-go response*), in different tasks. The same tones could also be irrelevant to making the appropriate behavioral outputs in the tasks. Neuronal activity evoked by the same tone differed when it was associated with an upcoming go or no-go response. The differences were observed in the spike activity of ~25% of the 420 multiunts and in the local field potentials at ~85% of the 570 sites recorded in core fields of auditory cortex. In both the spike activity and local field potentials, such differences emerged mainly because of increased activity evoked by

the tone when it was associated with a no-go response and barely changed activity when it was associated with a go response, relative to that when it was irrelevant to making an appropriate behavioral output. Our results suggest that tone representations in core fields of auditory cortex can change to reflect whether an upcoming go or no-go response should be made to the tones, i.e., auditory cortex can attach behavioral meaning to the tones.

Disclosures: Y. Huang: None. **M. Brosch:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.09/HH12

Topic: D.06. Audition

Support: NIH/NIDCD Grant F32C015160

NIH/NIDCD Grant R01C014452-01A1

Title: Maladaptive central auditory gain enhancement and disrupted loudness perception following acoustic trauma

Authors: *B. D. AUERBACH[1], K. RADZIWON[1], G.-D. CHEN[1], M. GHOBADI[2], E. T. ESFAHANI[2], R. SALVI[1]
[1]Ctr. for Hearing and Deafness, [2]Mechanical and Aerospace Engin., Univ. at Buffalo, Buffalo, NY

Abstract: The central auditory system displays a remarkable ability to adapt its response properties to changes in sound level. This includes compensatory increases in neuronal gain that can partially restore sound detection following long-term hearing loss. A price of this plasticity, however, is the potential for maladaptive sound encoding in response to abrupt or extreme changes in auditory input. To explore this issue, we examined the relationship between central auditory gain changes and behavioral measures of loudness following acoustic trauma. Simultaneous recordings from multiple levels of the central auditory system demonstrated that noise-induced gain enhancement gradually emerged along the ascending auditory pathway, culminating in excessive amplification of sound-evoked activity in the auditory cortex. In chronic recordings from behaviorally trained animals, we found this cortical hyperactivity to be strikingly correlated with changes in loudness perception. These results show that sound-evoked activity undergoes progressive amplification at successive stages of auditory processing following hearing loss and that excessive neural gain can result in maladaptive changes to sound intensity coding and loudness perception. These results have particular relevance to hyperacusis,

a common auditory perceptual disorder associated with hearing loss where moderate, everyday sounds are perceived as intolerably loud or painful.

Disclosures: B.D. auerbach: None. **K. Radziwon:** None. **G. Chen:** None. **M. Ghobadi:** None. **E.T. Esfahani:** None. **R. Salvi:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.10/HH13

Topic: D.01. Sensory Disorders

Support: ANR-12-JSV1-0008-01

Title: ER-mitochondria crosstalk is regulated by NCS and is impaired in Wolfram syndrome

Authors: *B. DELPRAT, C. DELETTRE
Inserm U1051, Montpellier, France

Abstract: Communication between endoplasmic reticulum (ER) and mitochondria plays a pivotal role in calcium (Ca^{2+}) signalling, energy metabolism and cell survival. Dysfunctions of this crosstalk lead to metabolic and neurodegenerative diseases. Wolfram syndrome (WS) is a fatal neurodegenerative disease due to mutations of the ER resident protein WFS1. However, clinical phenotype of WS resembles mitochondrial disorders. Here we show that WFS1 forms a complex with a Neuronal Calcium Sensor protein (NCS) and inositol 1,4,5-trisphosphate receptor (IP3R) to promote ER-mitochondrial Ca^{2+} transfer in response to stimuli that generate inositol-1,4,5-trisphosphate. In addition, we report that NCS localizes to mitochondria-associated membranes and regulates mitochondrial respiratory chain. Moreover, we demonstrate that WFS1 prevents the degradation of NCS by the proteasome. In agreement, NCS expression is reduced in WFS1-null patient fibroblasts, which show reduced ER-mitochondria interactions and Ca^{2+} exchanges. Importantly, NCS overexpression not only reactivates ER-mitochondrial Ca^{2+} transfer, but also induces a significant rescue of the dysfunctional mitochondrial phenotype observed in WFS1 deficient cells. Our results describe an unexpected key role of NCS in ER-mitochondria crosstalk and reconcile the ER expression of WFS1 with the mitochondrial phenotype, underlining a novel pathogenic mechanism for WS and opening new insights into the biogenesis of other neurodegenerative diseases.

Disclosures: B. Delprat: None. **C. Delettre:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.11/HH14

Topic: D.01. Sensory Disorders

Support: CAPES/PROAP

Title: Dyslexia: Evidence for a multifactorial basis

Authors: *M. G. FEITOSA[1], M. R. D. PRESTES, 70910-900[2], M. M. F. SANTANA, 70910-900[3,4], N. G. S. N. RIBEIRO[1]
[1]Inst. de Psicologia, Univ. De Brasília, BRASÍLIA, Brazil; [2]Hlth. Dept., Governo do Distrito Federal, BRASÍLIA, Brazil; [3]Ctr. Universitário do Distrito Federal, BRASÍLIA, Brazil; [4]Brasilia Children's Hosp., Brasília, Brazil

Abstract: According to the Phonological Theory, phonological deficit is a direct cause of dyslexia. According to the Auditory Deficit Theory, a change in the processing of acoustic stimuli is a direct cause of the change in the course of phonological development in dyslexics and, in turn, of the difficulty in learning to read and write. Since the speech stimulus is an acoustic signal, alteration in auditory temporal processing may lead to difficulty in processing of consonants characterized by rapid formant transitions. Alteration in perception of short sounds and rapid transitions can lead to difficulties in speech perception, impairing the construction of the mental representations of speech stimuli. Studies on categorical perception of speech in a voiced /unvoiced (V/U) perceptual continuum have shown altered perception of speech by dyslexics. And studies on the frequency of different types of orthographic errors have pointed out that voiced/voiceless exchanges are the most frequent type of error in both dyslexics and oralized deafs. Objective: To assess the underlying picture of the voiced/voiceless exchanges presented by dyslexics, and possible relationships between dyslexia symptomatology and alterations in auditory processing and speech perception. Methods: A group of Brazilian typical readers aged 9 to 15 years (N = 17), and two groups of dyslexics with (N = 17) and without (N = 09) V/U exchanges were studied. Groups were compared regarding measures of reading, writing, phonological awareness, minimum-pairs auditory discrimination, auditory temporal processing, and speech perception in an experiment of identification of stimuli that differed in onset time, forming the Portuguese perceptual continuum /bala-pala/. Results: The group of dyslexics without V/U exchanges did not differ from the group of typical readers in the occurrence of V/U exchanges, and in the total number of errors in the dictation task. In the other measures, the typical reader group presented superior performance to that of both groups of dyslexics. The groups of dyslexics with and without persistent V/U exchanges differed in the ability of auditory temporal resolution and in phonological awareness at the syllable level. Significant correlations

were observed between consistency in the classification of stimuli along the /bala-pala/ continuum and measures of reading, writing, phonological awareness, and auditory temporal processing. Conclusion: Evidence supports the proposition that dyslexia has a multifactorial basis, since both the auditory perceptual alteration and the phonological awareness alteration exerted influence on the symptoms of dyslexia.

Disclosures: M.G. Feitosa: None. **M.R.D. Prestes:** None. **M.M.F. Santana:** None. **N.G.S.N. Ribeiro:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.12/HH15

Topic: D.01. Sensory Disorders

Support: Evelyn Hart Watson Foundation

Title: Gata3 haploinsufficiency causes sensitivity to noise damage

Authors: *J. DUNCAN
Western Michigan Univ., Salt Lake City, UT

Abstract: Human *GATA3* haploinsufficiency leads to HDR (hypoparathyroidism, deafness, and renal dysplasia) syndrome. The development of the parathyroid, inner ear, and kidneys in humans appear extremely sensitive to levels of GATA3 and deviation of these levels leads to defects. Deafness in these patients appears to be moderate during childhood and progresses to severe in most patients. There has been some indication that in older mice with *Gata3* mutations there is a loss of inner ear hair cells while other auditory areas of the CNS appear normal. *Gata3* haploinsufficient mice were found to have relatively normal inner ear innervation, as well as normal innervation of the auditory and vestibular nucleus by the vestibular cochlear nerve. These mice also appear to have relatively normal hair cell and supporting cell development in the organ of Corti at birth. While these mice have normal numbers of cells and morphology of the organ of Corti and cochlear duct hearing was also tested by ABR. At 21 days mice had moderate hearing loss that continued throughout the lifespan. However when exposed to prolonged loud broad frequency sound the haploinsufficient mice had profound hearing loss and hearing did not return to normal. This is in contrast to control mice exposed to the same broad frequency sound. The control mice had a slight drop in hearing threshold that quickly returned to normal. While mice haploinsufficient for *Gata3* have moderate hearing loss, their hearing is quickly and permanently damaged by loud noise.

Disclosures: J. Duncan: None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.13/HH16

Topic: D.06. Audition

Support: U.S. Army Research Laboratory Grant W911NF-10-2-0022

U.S. Army Research Office Grant W911NF-14-1-0491

NINDS Grant NS040596

David M. Rubenstein Fund for Hearing Research

Title: Cortical auditory response adaptation and cross-frequency coupling in normal and impaired listeners

Authors: *U. MALINOWSKA[1], P. J. FRANASZCZUK[3], N. E. CRONE[4], D. F. BOATMAN[2]
[1]Neurol., [2]Johns Hopkins Sch. of Med., Baltimore, MD; [3]Human Res. & Engin. Directorate, US Army Res. Lab., Aberdeen Proving Ground, MD; [4]Neurol., Johns Hopkins Hosp., Baltimore, MD

Abstract: Introduction. In auditory cortex, the rapid decrease in neural activity that occurs with stimulus repetition is known as adaptation and is thought to facilitate listening in background noise. Stimulus repetition also increases cross-frequency coupling in auditory cortex, potentially reflecting sensory gating at the network level. We hypothesized that rate of adaptation and cross-frequency coupling correlate with listening performance in background noise.
Methods. We analyzed electrocorticographic (ECoG) recordings from 10 adult epilepsy patients with electrodes implanted over lateral, left temporal cortex. All had normal hearing; five demonstrated impaired speech recognition in noise. Tone and speech stimuli were presented in a passive auditory oddball paradigm. Time-frequency matching pursuit analysis was used to identify sites with significant increases in high-gamma (70-150 Hz; HG) power. Adaptation rates were computed for frequently repeated stimuli, fitting single-trial responses with a decaying exponential to derive time constants. Cross-frequency phase-amplitude coupling (PAC) of theta (4-7 Hz) and HG was measured by computing phase-locking values across consecutive repetitive trials. PAC changes as a function of stimulus repetition were identified by Pearson correlation coefficients.
Results. Patients with intact speech in noise abilities showed adaptation of high gamma power for repetitive tone and speech stimuli. Average adaptation rates were 2.23 repetitions for tones (0.8-5.75/patient) and 2.38 repetitions for speech (1.06-6.44/patient). For normal listeners, correlation analysis also revealed significant repetition-related increases in PAC for tones

(r=0.78, p=0.02) and even stronger increases for speech (r=0.9, p=0.002). For patients with impaired speech in noise listening abilities, the average HG adaptation rate for tones was 1.88 repetitions (0.8-10.5/patient), comparable to normal listeners. However, the average adaptation rate for speech was 4.02 repetitions (3.01-6.99/patient) almost twice that of normal listeners. Impaired listeners also showed no significant increases in PAC for tones (r= 0.06, p=0.98) or speech (r=-0.44, p=0.27).
Conclusions. In contrast to normal listeners, impaired listeners showed slower adaptation rates and no repetition-related increases in cross-frequency coupling in auditory cortex for tones or speech. These results suggest that slower adaptation and abnormal local network processing may contribute to listening difficulties in background noise.

Disclosures: U. Malinowska: None. **P.J. Franaszczuk:** None. **N.E. Crone:** None. **D.F. Boatman:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.14/HH17

Topic: D.06. Audition

Support: U.S. Army Research Office Grant W911NF-14-1-0491

U.S. Army Research Laboratory Grant W911NF-10-2-0022

Title: Effects of short term synaptic plasticity on stimulus specific adaptation in auditory cortex: A modeling study

Authors: *D. BEEMAN[1], P. KUDELA[2], D. BOATMAN-REICH[3], W. S. ANDERSON[2]
[1]Univ. of Colorado Boulder, Boulder, CO; [2]Neurosurg., Johns Hopkins Univ., Baltimore, MD; [3]Neurol. and Otolaryngology, Johns Hopkins Sch. of Med., Baltimore, MD

Abstract: Neural responses decrease with stimulus repetition, a phenomenon known as adaptation or repetition suppression. In auditory cortex, adaptation is stimulus-specific (SSA) in that it does not generalize to novel or different sounds and is often measured using an 'oddball paradigm' in which the subject listens to a weighted random sequence of short tone pips at two different frequencies, with one more probable than the other. Auditory evoked potentials (AEPs) are recorded from cortical surface (ECoG) electrodes.
The neural mechanisms underlying SSA in human auditory cortex remain poorly understood. In this modeling study, short term synaptic depression and facilitation were evaluated as a potential neural substrate for SSA in auditory cortex. Model simulations based on the same experimental parameters as the ECoG recordings yielded AEPs consistent with those derived from the ECoG

recordings.
An existing layer 4 primary auditory cortex model consisting of 2304 multicompartmental pyramidal cells and 576 basket cells was extended to include 288 inhibitory Martinotti cells. Synaptic depression was applied to interlayer excitatory synapses of the pyramidal and basket cells, with facilitating excitatory synapses on the Martinotti cells.
SSA arises from two time constants associated with the plasticity model, which was implemented in GENESIS 2.4. After a fit of the model parameters to measurements in mouse auditory and somatosensory cortex, we were able to replicate and explain the significant features of the AEP experiments.
As in a typical experiment or simulation, 300 short tone pips at 1000 and 1200 Hz are presented at 1 sec intervals with probabilities of 0.82 and 0.18. A thalamic input model applies tones targeted at rows along a tonotopic axis with a 1 mm spacing between octaves. Thus, two mostly distinct groups of neurons are strongly excited. On average, frequent tone stimuli will follow each other at 1 sec intervals. The probability of two sequential infrequent tones is low, and the average interval between these stimuli is much larger.
In the synaptic depression model, the synaptic weight is a product of two depression factors, each with a depression increment and a time constant for weight decay back towards the original value of 1.0. One of these, with a time constant of about 1.5 sec, gives less depression if the interval between input spike pulses is long. Consequently, sequential stimuli of the frequent tone will maintain some depression between pulses, but responses to the more widely separated infrequent stimuli will have recovered from the depression. This is seen in the amplitudes of simulated AEPs for frequent and infrequent tones.

Disclosures: **D. Beeman:** None. **P. Kudela:** None. **D. Boatman-Reich:** None. **W.S. Anderson:** F. Consulting Fees (e.g., advisory boards); Longeviti, LLC - compensated advisory board member, Globus Medical - compensated consultant.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.15/HH18

Topic: H.01. Animal Cognition and Behavior

Support: NIH Grant MH46904

NIH Grant MH74006

NIH Grant T32 MH106454

Title: Mechanistic identification of stimulus relay from the medial auditory thalamus necessary for delay and trace eyelid conditioning

Authors: *L. C. HOFFMANN, S. J. ZARA, M. D. MAUK
Ctr. for Learning and Memory, Univ. of Texas At Austin, Austin, TX

Abstract: The essence of neural processing is transforming inputs into useful output. Identifying and characterizing inputs is therefore a fundamental initial step toward understanding any particular instance of neural processing. How discrete, stimulus-driven responses are transformed into persistent neural responses that last well beyond stimulus offset, a process often connected to the substrates of working memory (Fuster & Alexander 1971; Funahashi, Bruce & Goldman-Rakic, 1989; Zhou & Fuster 1996), is an important example. Trace eyelid conditioning requires persistent activity that is conveyed to the cerebellum for learning to occur (Kalmbach et al. 2009; Kalmbach, Ohyama & Mauk 2010). To begin identifying where and how this persistent activity is generated, we have tested the hypothesis that the medial auditory thalamic nuclei (MATN) are a necessary component of a pathway connecting the activity elicited by auditory conditioned stimuli (CS) to the task-related persistent activity that is necessary for learning. Using tetrode recordings of medial prefrontal cortex and reversible MATN inactivation with muscimol, we show that the MATN are necessary for trace ELC and corresponding persistent activity in prefrontal regions necessary for the task. Using electrical stimulation of MATN, we further show that CS-duration input from MATN is not sufficient for acquisition of trace conditioning, while inputs that bridge CS and US support learning. Results suggest trace eyelid conditioning is supported through persistent activity that is driven directly or indirectly by MATN.

Disclosures: L.C. Hoffmann: None. **S.J. Zara:** None. **M.D. Mauk:** None.

Poster

400. Auditory System Plasticity

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 400.16/HH19

Topic: D.01. Sensory Disorders

Title: Hearing sensitivity and participation restriction in adults with longstanding unilateral chronic otitis media

Authors: *R. S. TSCHIEDEL[1], S. C. L. BRAGA[2], L. S. SÁ[2], M. V. S. MEDEIROS[1], R. C. GRANJEIRO[2], M. A. G. FEITOSA[1]
[1]Univ. of Brasilia, Brasilia, Brazil; [2]Distrito Federal Dept. of Hlth., Brasilia, Brazil

Abstract: Chronic otitis media (COM), a characteristic disease of developing countries, can cause a decrease in auditory sensitivity. This perceptual barrier, often initiated in childhood and adolescence, can extend throughout adult life due to several factors. Hearing impairment resulting from COM can lead to negative implications for daily activities that demand auditory perception, even when the involvement is unilateral. The objective of this study was to assess the

level of auditory sensitivity, perceived hearing difficulty, and auditory participation restriction among adults with a longstanding unilateral COM history. Twenty-six adults with unilateral COM, whose symptoms started until the age of 18, were evaluated. All were followed up at a public Otorhinolaryngology outpatient clinic in the Brazilian capital and were recruited from the waiting list for otologic surgery. Procedures included interview, puretone thresholds for 500, 1000, 2000 and 4000 Hz, and administration of the Hearing Handicap Inventory for Adults questionnaire (score 0 to 100).In the sample aged 18-54 years, 54% had the symptoms of COM initiated until 6 years of age, and 46% between 7 and 18 years of age. These participants had on average symptoms of COM for 33 years, and 65% of them had undergone at least one otologic surgery. In all, the ear with COM presented alteration in the peripheral auditory system, and in 69% there was involvement of the inner ear associated with alteration in the middle ear, and in 99% reduction in the auditory sensitivity (mean = 46dB). Sixty-five percent presented some level of participation restriction (HHIA ranged from 18 to 80), and for 69% the perceived hearing difficulty was classified as medium or large. Both, participants with a mean auditory sensibility threshold between 21 and 40dB, and participants with an average threshold between 41 and 69dB, presented levels of restriction of absent to severe participation. Correlation analysis using Kendal tau showed that perceived hearing difficulty correlated with HHIA score ($\tau = 0.34$, $p < 0.05$) and the consequent level of participation restriction ($\tau = 0.83$, $p < 0.01$). Results also showed that HHIA score cannot be explained based solely on auditory sensitivity, and other factors should be investigated. Adults with unilateral COM may present a long time of hearing deprivation, even though they have undergone otologic surgery, and each individual perceives differently the reduction in auditory sensitivity.

Disclosures: R.S. Tschiedel: None. **S.C.L. Braga:** None. **L.S. Sá:** None. **M.V.S. Medeiros:** None. **R.C. Granjeiro:** None. **M.A.G. Feitosa:** None.

Poster

401. Retina: Motion

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 401.01/HH20

Topic: D.07. Vision

Support: HHMI

Title: Selective synaptic contacts promote retinotopic detection of visual looming in the *Drosophila* central brain

Authors: *M. B. REISER, M. MORIMOTO, A. NERN, G. M. RUBIN, E. ROGERS, A. WONG, P. GHORBANI, N. A. SMITH, M. DREHER, R. PAREKH, D. BOCK
HHMI / Janelia, Ashburn, VA

Abstract: Lobula Columnar (LC) neurons provide an anatomical connection between early visual processing areas and central brain regions in flies. Each LC cell class projects to distinct central brain structures called optic glomeruli. LC neurons exhibit retinotopic tiling of a visual neuropil with their dendrites, but their axonal projections in the glomeruli do not show any obvious retinotopic organization. These cells have been thought to encode visual features, and this apparent loss of retinotopy provided further evidence for this proposal. We have focused on a single LC type, the LC6 neurons, which are responsive to visual looming stimuli, and can induce takeoff behavior in flies upon ontogenetic activation. Since the LC6 glomerulus is not innervated by the known neurons of escape pathways, we investigated the targets of LC6 projections. Using the imaged expression pattern of collections of genetic driver lines we have identified at least 5 downstream target neuron types of LC6, and then used the split-GAL4 technique to develop refined genetic driver lines for each cell type. We established the functional connectivity of LC6s to each downstream neuron type by using optogenetic depolarization of LC6 neurons while measuring intracellular calcium responses in the downstream neurons. We then used 2-photon calcium imaging to measure the visual response properties of several LC6 target neurons. While the target neurons retain the sensitivity to visual looming, we find that different downstream cell types appear to respond selectively to looms presented in distinct, restricted, parts of the visual field. In light of the previous evidence observed with light microscopy, that the optic glomerulus does not contain any retinotopic organization, these new imaging results suggested that retinotopy might be achieved through selective synaptic contacts in the glomerulus. To examine this hypothesis, we took advantage of the Bock Lab's newly acquired serial section TEM imaging volume of an entire female Drosophila brain. We began tracing visual projections into the central brain and were able to identify the LC6 neurons as well as their targets. Several target neurons and all of their LC6 inputs were traced. We observe selective connectivity between the target neurons and their visual inputs, suggesting that distinct patterns of visual information are accessible within optic glomeruli.

Disclosures: **M.B. Reiser:** None. **M. Morimoto:** None. **A. Nern:** None. **G.M. Rubin:** None. **E. Rogers:** None. **A. Wong:** None. **P. Ghorbani:** None. **N.A. Smith:** None. **M. Dreher:** None. **R. Parekh:** None. **D. Bock:** None.

Poster

401. Retina: Motion

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 401.02/HH21

Topic: D.07. Vision

Support: HHMI support through Michael Reiser

Title: A visual projection neuron class links the detection of translational motion to the control of forward walking

Authors: *M. ISAACSON, J. ELIASON, A. NERN, M. B. REISER
Janelia Res. Campus, HHMI, Ashburn, VA

Abstract: The perception of visual motion is useful for animal navigation and moving object detection, and flies are a prominent model system for elucidating the neural mechanisms behind this computation. In flies, directionally selective neurons in the optic lobe compute local motion and project retinotopically into motion-direction-specific layers in the Lobula Plate. Several large visual projection neurons have been extensively studied that integrate Lobula Plate local motion information across a large field-of-view of the fly eye and project this information to the central brain. However, the Drosophila visual system also features small-field visual-projection neurons which have received little attention. Here we present the results of studying one such cell type, which we call Lobula Plate Columnar Type 1 (LPC1), in tethered flies positioned in the center of an LED visual arena capable of delivering all types of rotational and translational motion stimuli. We found that regressive translational motion, which specifically caused flies to cease forward walking, no longer had this effect upon silencing LPC1, yet LPC1 silencing did not affect optomotor reactions to rotational visual motion. We then used 2-Photon imaging to measure visual stimulus evoked calcium responses in the axon terminals of LPC1s, and found that this cell type responds strongly to regressive motion presented to its corresponding eye, but this response is abolished when the opposite eye simultaneously received progressive motion -- a computation that can differentiate between the visual consequence of translational motion and rotational self-motion. Optogenetic activation of LPC1 caused the fly to cease forward walking while also not affecting turning behaviors produced by rotational motion stimuli, demonstrating that the control of forward locomotion and turning are largely decoupled in the fly nervous system. These results identify a population of cells that integrate visual translational motion information from both eyes, a computation that may contribute to the behavioral reactions to moving objects.

Disclosures: **M. Isaacson:** A. Employment/Salary (full or part-time):; HHMI - Janelia Research Campus. **J. Eliason:** A. Employment/Salary (full or part-time):; HHMI - Janelia Research Campus. **A. Nern:** A. Employment/Salary (full or part-time):; HHMI - Janelia Research Campus. **M.B. Reiser:** A. Employment/Salary (full or part-time):; HHMI - Janelia Research Campus.

Poster

401. Retina: Motion

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 401.03/HH22

Topic: D.07. Vision

Support: HHMI

Title: Simple integration of inputs computes directional selectivity in *Drosophila*

Authors: *E. GRUNTMAN[1], S. ROMANI[2], M. B. REISER[3]
[1]Janelia Res. Campus, Ashburn, VA; [2]HHMI Janelia Res. Campus, Ashburn, VA; [3]HHMI Janelia Res. Campus, Ashbrun, VA

Abstract: The detection of visual motion is a fundamental neuronal computation that serves many critical behavioral roles, such as encoding of self-motion or figure-ground discrimination. For a neuron to extract directionally selective (DS) motion information from inputs that are not motion selective it is essential to integrate across multiple spatially distinct inputs. This integration step has been studied for decades in both vertebrate and invertebrate visual systems and given rise to several competing computational models. Recent studies in Drosophila have identified the 4th-order neurons, T4 and T5, as the first neurons to show directional selectivity. Due to the small size of these neurons, recordings have been restricted to the use of calcium imaging, limiting timescale and direct measurement of inhibition. These limitations may prevent a clear demonstration of the neuronal computation underlying DS, since it may depend on millisecond-timescale interactions and the integration of excitatory and inhibitory signals. In this study, we use whole cell in-vivo recordings and customized visual stimuli to examine the emergence of DS in T4 cells. We record responses both to a moving bar stimulus and to its components: single position bar flashes. Our results show that T4 cells receive both excitatory and inhibitory inputs, as predicted by a classic circuit model for motion detection. Furthermore, we show that by implementing a passive compartment model of a T4 cell, we can account not only for the DS response of the cell, but also for its dynamics.

Disclosures: E. Gruntman: None. **S. Romani:** None. **M.B. Reiser:** None.

Poster

401. Retina: Motion

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 401.04/HH23

Topic: D.07. Vision

Support: BB/MO12093/1

Title: Circuitry of the lobula giant movement detector 2 neuron

Authors: *E. COCKS, C. RIND
Newcastle Univ., Newcastle Upon Tyne, United Kingdom

Abstract: Serial Block Face Scanning Electron Microscopy (SBF-SEM) can reveal novel information on neuronal circuitry. One example is the discovery of the complex circuitry of the locust visual system, specifically the Lobula Giant Movement Detector 1 and 2 (LGMD1 and 2) neurons. The neurons are part of a larger network involved in jumping, escaping and hiding in response to an approaching object. The system is of importance, as it can be used as a model of sensory-motor integration. However, the LGMD1 has been the main focus of analysis so far and not much is known about the connectomics of the LGMD2. In order to study this with SBF-SEM, the brain of an adult locust (*Locusta migratoria*) was processed with an adapted heavy metal staining protocol. The samples were embedded in hard resin and the blocks trimmed and prepared for SBF-SEM. From a block 500 slices at 60nm thickness were taken and analysed in Microscopy Image Browser and Amira. Reconstructions of the LGMD2 dendrites show single afferent neurons (AN) from the medulla, connecting with the LGMD2 more than once. ANs each synapse onto both the large (~7µm) and small (~2µm) diameter branches, in a ~18-20µm area. ANs synapsed multiple times with the same and different neighbouring ANs at each of these two locations. Also seen, and only through 3D reconstruction, is that the ANs, found to synapse multiple times in one region on the LGMD2, followed a similar path to one another through the neuropil, when no longer synapsing. ANs have previously been shown to synapse on multiple occasions on the same branch, and on multiple branches of the LGMD1. The reconstructions show a similar pattern for the LGMD2. Multiple synaptic connections in a small area of the LGMD2 from a single AN allows for rapid signal integration over the LGMD2 dendrites, explaining how a locust can process a visual stimulus and initiate a motor response in less than a tenth of a second. The precise computation of looming by the LGMD1 relies on the multiplication, in the neuron, of two signals, one excitatory and the other inhibitory. The inhibitory signal is spatially removed from the excitatory one and this matches the structure of the LGMD1 dendritic tree, which has one main dendritic tree for excitatory input (branch A) and two smaller dendritic fields (sub-branches B and C) for inhibitory input. The LGMD2 does not have these sub-branches so how is looming signalled? Labelling of inhibitory synapses in the neuropil, using immunogold, in combination with SBF-SEM, could be a way to find and map another mechanism of inhibition that occurs in the input network of the LGMD2. Revealing how the combination of excitatory and inhibitory inputs also shapes the LGMD2's response to looming objects.

Disclosures: **E. Cocks:** None. **C. Rind:** None.

Poster

401. Retina: Motion

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 401.05/HH24

Topic: D.07. Vision

Title: Electron microscopy-based reconstruction of the motion information processing circuits in the fruit fly brain

Authors: *K. SHINOMIYA[1], G. HUANG[1], T. ZHAO[1], S. XU[2], S. PLAZA[1], L. SCHEFFER[1], I. A. MEINERTZHAGEN[3,1]
[1]FlyEM Project Team, [2]HHMI Janelia Res. Campus, Ashburn, VA; [3]Dept. of Psychology & Neurosci., Dalhousie Univ., Halifax, NS, Canada

Abstract: The visual system of the brain of *Drosophila melanogaster* has been studied as a model system in the field of electron microscopy (EM)-based connectomics. Regardless of its modest size, the optic lobe houses complex circuitry, which processes diverse modalities of visual information in parallel. In this study, we reconstruct motion information processing circuits in the optic lobe applying a sparse reconstruction strategy using the focused-ion beam aided scanning EM (FIB-SEM) technique for imaging. The dataset covers the entire optic lobe, including three synaptic neuropils, the medulla, lobula and lobula plate. The neuropils, consisting retinotopic columnar units, are interconnected by neuronal processes through the second optic chiasm. We imaged the optic lobe neuropils with FIB-SEM with an isotropic resolution (8nm x 8nm x 8nm). Neurons in the three-dimensional grayscale image stack were segmented on computer, and synaptic areas (both pre- and post-synapses) were also auto-predicted. It has been known that motion information is detected by T4 cells projecting from the medulla to the lobula plate, and T5 cells projecting from the lobula to the lobula plate. T4 and T5 cells detect moving light (ON)- and dark (OFF)-edges respectively, with inputs from their upstream neurons. Motion information from the two pathways is integrated into lobula plate neurons. Using information of predicted synapses, we traced and reconstructed neurons that receive inputs in the lobula plate to comprehensively identify the neurons participating in motion information processing. As a result, we reconstructed the entire motion information processing circuit in the optic lobe completely from the photoreceptors to the projection neurons to the central part of the brain. We confirmed that many lobula plate tangential cells (LPTCs), lobula plate interneurons (LPi) and other newly found neurons are engaged in motion information processing, with projections from the T4 and T5 cells. In the lobula plate, four directions of stationary motion (backward, forward, upward and downward) are processed in four distinct layers. We also revealed that T4 and T5 cells processing motion of one direction inhibit LPTCs responsible for the opposite direction in a neighboring layer, mediated by inhibitory LPi cells. This connectomics approach is expected to accelerate high-throughput reconstruction of neuronal circuits that serve as the anatomical basis for further functional analyses, both within the optic lobe and in the central brain.

Disclosures: **K. Shinomiya:** None. **G. Huang:** None. **T. Zhao:** None. **S. Xu:** None. **S. Plaza:** None. **L. Scheffer:** None. **I.A. Meinertzhagen:** None.

Poster

401. Retina: Motion

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 401.06/HH25

Topic: D.07. Vision

Support: Howard Hughes Medical Institute

Title: Comprehensive connectome of the fly's visual and olfactory circuits using FIBSEM

Authors: *S.-Y. TAKEMURA[1], S. PLAZA[2], L. SCHEFFER[3], I. A. MEINERTZHAGEN[4]
[1]Janelia Res. Campus/HHMI, Ashburn, VA; [2]Fly EM Team, HHMI, Ashburn, VA; [3]Howard Hughes Med. Inst. Janelia Farm Res. Campus, Ashburn, VA; [4]Dept. of Psychology & Neurosci., Dalhousie Univ., Halifax, NS, Canada

Abstract: A classic problem in visual neuroscience, how the visual system detects image motion, has recently undergone resurgent interest in the fruit fly Drosophila. The problem has long been analyzed using mathematical models, of which the Hassenstein-Reichardt elementary motion detector (EMD) or Barlow-Levick model are both widely canvassed. Both models compare two visual inputs separated in time and space and simulate motion detection well. However, the exact neural mechanism underlying the detection of direction by motion-sensing cells is still understood only imperfectly. Here we reconstructed a comprehensive connectome of the circuits of fly's visual motion-sensing T4 cells using a superior electron microscopy technique. We uncover complex T4 inputs and reveal that putative excitatory inputs cluster at T4's dendrite shafts, while inhibitory inputs localize to the bases. Our results suggest new candidate circuits for motion detection, including previously unidentified inhibitory pathways. We furthermore mapped synaptic circuits of ~30,000 connections between nearly all medulla cell types, and also reconstructed the morphologies of, and mapped over 240,000 synaptic connections between, all 983 neurons within the mushroom body, the fly's learning and memory center. The unprecedented level of detail of synaptic circuits should enable modeling studies not previously possible and suggests many experiments to explore their physiological and behavioral significance. That many of the new pathways and neural circuitry were not anticipated by decades of extensive anatomical, experimental and theoretical studies argues strongly for the value of EM connectomics studies.

Disclosures: S. Takemura: None. **S. Plaza:** None. **L. Scheffer:** None. **I.A. Meinertzhagen:** None.

Poster

401. Retina: Motion

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 401.07/HH26

Topic: D.07. Vision

Title: The effect of locomotion-induced octopamine release on motion detection circuits in *Drosophila*

Authors: *J. KOHN, R. BEHNIA
Columbia Univ., New York, NY

Abstract: To survive in ever-changing environments, an animal must be able to detect relevant stimuli. Neuromodulation affords anatomically rigid networks the ability to tune their sensitivity to various stimuli in a state-dependent manner. However, the mechanisms underlying the modulation of neural circuits are relatively unknown. Motion-detecting circuits in *Drosophila* are well-characterized and provide a powerful model system to study the effects of behavioral state on circuit output. These circuits consist of connections between photoreceptors in the eye and lobula plate tangential cells (LPTCs) in the optic lobe, which are direction selective. Locomotion modulates LPTCs by increasing their response amplitude and shifting their sensitivity toward faster moving stimuli (Maimon *et al.* 2010, Chiappe *et al.* 2010). This process is dependent upon the release of octopamine (OA), a neuromodulator equivalent to mammalian norepinephrine (Suver *et al.* 2012). We investigate the neuronal targets of OA in these circuits, how the action of OA leads to the shift in sensitivity of the outputs towards faster-moving stimuli, and explore potential cellular and circuit mechanisms underlying this modulation.

Disclosures: J. Kohn: None. **R. Behnia:** None.

Poster

401. Retina: Motion

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 401.08/HH27

Topic: D.07. Vision

Support: HHMI Janelia Research Campus

Title: Flight control and color vision in *Drosophila*

Authors: *K. D. LONGDEN, M. B. REISER
HHMI Janelia Res. Campus, Ashburn, VA

Abstract: Learning and memory experiments have demonstrated that *Drosophila* has color vision and can discriminate different wavelengths of light, even when they are isoluminant. This capacity for color vision is supported by the expression of five rhodopsins in the compound eyes, each with a distinct wavelength sensitivity profile, and by dedicated circuitry that has been characterized in detail. Meanwhile, how naïve flies use for this capacity for color vision remains a mystery. Here, we have investigated how color vision can influence three forms of flight control prior to learning. To do this, we developed a novel ultraviolet and green projector system to display wide-field visual stimuli. We measured the flight control responses of tethered flies by optically recording changes in wing stroke amplitude. First, flies can stabilize the horizon even when the intensities of the different wavelengths are matched so that they provide no luminance contrast. This is achieved by combining wavelength-sensitive phototaxis and color-blind motion vision to stabilize the scene. Second, during looms the steering responses are color-blind, but the wing beat frequency varies with the intensity of ultraviolet light. Third, during flight towards an attractive object, a single vertical stripe, the responses are color blind. Together, our results show how the wavelength of light can influence multiple aspects of flight attitude and control, and allow the operation of color vision circuitry to be investigated in the context of natural behaviors.

Disclosures: K.D. Longden: None. **M.B. Reiser:** None.

Poster

401. Retina: Motion

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 401.09/HH28

Topic: D.07. Vision

Support: NIH MH065339,

NSF DMS-1120952

Title: Characterization of feed forward inhibition in a looming detection circuit

Authors: *H. WANG[1], R. B. DEWELL[1], F. GABBIANI[1,2]
[1]Neurosci., Baylor Col. of Med., Houston, TX; [2]Dept. of Electrical Engin., Rice Univ., Houston, TX

Abstract: The lobula giant movement detector (LGMD) in the locust optic lobe is among the best-studied looming sensitive neurons. It receives feed-forward excitatory inputs through a fan-shaped dendritic field, called field A and inhibitory inputs through two additional dendritic fields, called B and C. The excitatory inputs encode the angular velocity of the approaching

object, while the inhibitory inputs are thought to encode its angular size. Current experimental data are consistent with the LGMD effectively multiplying these two inputs resulting in a firing rate that gradually increases, peaks, and then decays towards collision. The medullary neurons providing inhibition to the LGMD have not yet been physiologically characterized. Hence, our current understanding of feed-forward inhibition is based on anatomy, pharmacology and modeling. Multiunit recordings from the medulla were paired with intracellular LGMD recordings during visual stimuli to characterize the inhibitory inputs to the LGMD. Previously, we examined their sensitivity to contrast polarity, directional selectivity, receptive field size and the response profile. In this work, the inhibitory neurotransmitters released onto dendritic field C were examined. To our surprise, puffing of multiple transmitters, including GABA and Histamine, significantly reduced the firing rate of the LGMD in response to looming stimuli. For the same stimuli, the recorded medullary neurons showed a firing rate profile resembling that of the LGMD, which gradually increases, peaks and then decays towards the end of the simulated approach. An angular size linear model fits the instantaneous firing rate of the recorded medullary neurons with a R^2 value of 0.71. This indicates that the firing rate of medullary neurons providing inhibitory input to the LGMD most likely encodes angular size of the approaching object, in agreement with the earlier hypothesis that a logarithmic exponential transform implements multiplication of angular size with a negative exponential of speed within the LGMD neuron.

Disclosures: H. Wang: None. **R.B. Dewell:** None. **F. Gabbiani:** None.

Poster

401. Retina: Motion

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 401.10/HH29

Topic: D.07. Vision

Support: NSF MRI-1626326 (VDCS)

Towson University Graduate Student Association Award (JS)

Title: Electroretinographic study of vision in the stink bug Halymorpha halys compound eyes

Authors: J. STRICKLAND[1], A. B. LALL[2], *V. D. SHIELDS[1]
[1]Fisher Col. of Sci. and Mathematics - Biol. Sci., Towson Univ., Towson, MD; [2]Biol., Howard Univ., Baltimore, MD

Abstract: The brown marmorated stink bug, *Halymorpha halys*, has become an agricultural nuisance in the United States. According to the Environmental Protection Agency, this polyphagous feeder creates damage and yield loss in a wide range of crops. The insect utilizes a

suckering mechanism for the removal of sap from desirable food sources. The affected crops are often left unappealing to consumers. The typical methods utilized for prevention of damage and ultimately loss have been non-biological pesticides, which are designed to kill or cause adverse effects to living organisms. However, more recent control measures have included the design of olfactory and visual traps. The incorporation of visual traps as an alternative to common non-biological pesticides are beneficial to both human health and other living organisms. To utilize visual traps, it is first necessary to know the visual characteristics of the compound eyes of this insect. Electroretinograms (ERGs) elicited by light stimuli were recorded from the corneal surface of the eyes by a standard method. Our results indicate that the ERG was a negative monophasic waveform consisting of an initial phasic component followed by a maintained component that lasted for the duration of the flash, with no off-response at the end of white light stimulus. The eye responded over a six log unit change in white light illumination, as indicated by the V/logI curves, and the response latency was long (40-60 ms) in dim illumination and short (20-30 ms) in bright illumination. Visual spectral sensitivity of the eye was obtained by determining the number of photons needed for eliciting a criterion amplitude ERG response (0.2, 1.2, and 2.4 mV) across the spectrum at 13 different stimulus wavelengths from 420 nm to 640 nm. The spectral sensitivity had a maximum around 540 nm. The waveform of the ERG response elicited by short wavelength stimuli (blue) differed from those obtained with long wavelength stimuli (green or red). These waveform responses are being further evaluated by studies obtaining ERG responses under different chromatic adaptation conditions. Our data suggest the presence of a dominant spectral mechanism in the green region of the spectrum as is common among insects. The presence of a secondary spectral mechanism in the short wavelengths is being investigated by chromatic adaptation experiments.

Disclosures: **J. Strickland:** None. **A.B. Lall:** None. **V.D. Shields:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.01/HH30

Topic: D.07. Vision

Support: NIH Grant EY05253 (JMA)

NIH Grant EY013312 (QZ)

Title: Linear relation between orientation structure and spatial resolution in visual cortex

Authors: *E. KOCH, J. JIN, J. M. ALONSO, Q. ZAIDI
Biol. and Visual Sci., SUNY Optometry, New York, NY

Abstract: Visual information is topographically mapped in the primary visual cortex according to multiple stimulus parameters such as orientation and spatial frequency. While the representation of each parameter is well known, the cross-parameter structure remains poorly understood. We recently proposed an Excitation-Normalization (EN) model to explain the topographic relation between orientation tuning and orientation preference (Koch et al, 2016). This model also predicts a strong relation between local orientation homogeneity and spatial frequency resolution. Here we tested the prediction for spatial resolution by performing horizontal penetrations with multielectrode arrays through cat primary visual cortex. We simultaneously measured orientation and spatial frequency tuning with random sequences of single gratings that included 8 orientations, 4 phases and 10 spatial frequencies presented separately to contralateral and ipsilateral eyes. For each cortical site, we calculated a homogeneity index of local orientation preference (LHI, 1: no local change, 0: rapid local change), the circular variance of orientation tuning, the high spatial frequency cutoff at 50% response amplitude (SF50), and the ocular dominance. As predicted by the EN model, our results revealed a strong linear relationship between SF50 and LHI ($r=0.7099$, $p<0.0001$, $n= 55$), with spatial resolution systematically increasing as the multielectrode entered a cortical iso-orientation domain. The SF50 was also correlated with orientation selectivity ($r=-0.5767$, $p<0.0001$, $n=55$) and ocular dominance ($r=0.4604$, $p<0.001$, $n=55$), with non-oriented monocular-neurons showing lower spatial resolution than orientation-selective binocular-neurons, as recently found in macaques (Nauhaus et al, 2016). We also found a strong relation between ocular dominance and binocular mismatch in spatial resolution ($r=0.7246$, $p<0.0001$, $n=45$), with neurons being binocularly-matched in spatial resolution when they are also binocularly-matched in response strength. Finally, ocular dominance was correlated with LHI ($r=-0.3313$, $p=0.003$, $n=75$) as expected from the alignment of pinwheel centers with monocular regions. Our results reveal pronounced relations across different stimulus parameters in cortical topography as would be expected from a specialization of different map domains in different functions. We conclude that, just as the weak cross-orientation suppression of pinwheel centers allow processing of multi-orientation patterns more efficiently (Koch et al, 2016), the enhanced spatial resolution of binocular domains may help maximize the spatial resolution of binocular vision.

Disclosures: E. Koch: None. **J. Jin:** None. **J.M. Alonso:** None. **Q. Zaidi:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.02/HH31

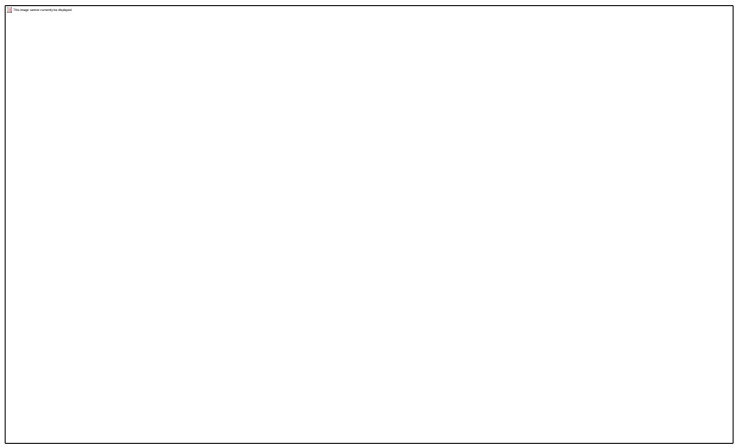
Topic: D.07. Vision

Support: NSCERC

Title: Emergent functional connectomes are systematically activated within overlapping micro-organization of spatial frequency and orientation maps in the visual cortex

Authors: *S. MOLOTCHNIKOFF[1], V. BHARMAURIA[2], N. CHANAURIA[3], L. BACHATENE[4]
[1]Sci. Biologiques, Univ. de Montreal, Montreal, QC, Canada; [2]York Univ., North York, ON, Canada; [3]Sci. Biologiques, Univ. de Montréal, Montréal, QC, Canada; [4]Dept. of Nuclear Med. and Radiobiology, Univ. de Sherbrooke, Sherbrooke, QC, Canada

Abstract: Orientation tuning and spatial frequency tuning are characteristic properties of visual neurons across species. Although the orientation maps are well established and investigated across species, debate still persists on the existence of the spatial frequency maps in the cortex. However, recently, investigators using optical imaging (Nauhaus et al. 2012; Ribot et al. 2013), two-photon imaging (Nauhaus et al. 2012) and single neuron electrophysiology (Molotchnikoff et al. 2007) have shown and suggested that spatial frequency is micro-organized and overlapped with orientation maps in the visual cortex. Here we investigated the functional circuits (connectomes) within ensembles of neurons (recorded using tungsten microelectrodes in anesthetized cats) while spatial frequency and orientation tunings of neurons were simultaneously tested. First, the orientation tunings of neurons were computed by randomly presenting oriented sine-wave drifting gratings in steps of 22.5°, and then the functional circuits were revealed using cross-correlograms (CCG) between these simultaneously recorded neurons at each orientation (Bharmauria et al. 2016). Thereafter, the spatial frequency was systematically changed and the functional connectomes were revealed within above ensembles while the orientation was kept constant (0° or 90°). Indeed, as reported before (Bharmauria et al. 2014, 2016), a salient network was activated within an ensemble in relation to the specific spatial frequency, suggesting that a unique functional connectome is important for spatial frequency encoding. Further, it was divulged that neurons that shared orientation and spatial frequency tuning exhibited significantly more connections than those neurons that did not share the properties. This suggests that spatial frequency and orientation tuning maps may be simultaneously micro-organized and functionally superimposed while an ensemble encodes both properties at the same time.
P.S. SM and VB contributed equally



Disclosures: **S. Molotchnikoff:** None. **V. Bharmauria:** None. **N. Chanauria:** None. **L. Bachatene:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.03/HH32

Topic: D.07. Vision

Support: NIH Grant EY05253

NIH Grant EY027157

Title: Functional specialization of ON and OFF pathways for slow-global and fast-local change in the visual world

Authors: *R. MAZADE, J. JIN, C. PONS, J. ALONSO
Biol. and Vision Sci., State Univ. of New York Col. of Optometry, New York, NY

Abstract: Visual information is processed in the brain by ON and OFF pathways that respond to light and dark stimuli. In the visual cortex, OFF visual responses are stronger, represent visual space more accurately, and respond more linearly to luminance contrast than ON responses. Here, we demonstrate that these ON-OFF cortical asymmetries reflect a functional specialization to signal different spatial (global vs. local) and temporal (slow vs. fast) properties of visual scenes. To quantify the ON/OFF cortical representation of spatiotemporal space, we performed horizontal penetrations through cat primary visual cortex with multielectrode arrays (32-electrode Neuronexus, 0.1 mm electrode spacing, n=5 animals) and measured visual responses to dark and light targets presented with different sizes (1 to 23 deg/side), durations (16 to 133 msec), backgrounds (light, dark, mid-gray) and mean luminance (0.024 to 239 cd/m^2, attenuated with neutral density filters). Our results demonstrate that cortical responses are 25% stronger to darks than lights when stimuli are fast and spatially local (e.g. 16 msec, 7 deg target; dark=106.5±3.5 vs. light=80.1±2.3 spk/s; $p<0.001$, Wilcoxon test) but they are 30% stronger to lights than darks when stimuli are slow and large (e.g. 133 msec, 23 deg target; light=85.4±2.1 vs. dark=60.5±1.6 spk/s; $p<0.001$, Wilcoxon test). In addition, we demonstrate that responses to darks show pronounced size suppression (e.g. 16 msec, 7 vs. 23 deg target; 106.5±3.5 vs. 85.1±2.4 spk/s; $p<0.001$, Wilcoxon tests) and temporal suppression (e.g. 7 deg target, 16 vs. 133 msec duration; 106.5±3.5 vs. 70.6±1.9 spk/s; $p<0.001$, Wilcoxon tests), two properties that are important to segregate local moving targets in visual scenes. Conversely, responses to lights are robust when the stimulus is large and show no size suppression (e.g. 7 vs. 23 deg target; 80.7±1.1 vs. 82.6±1.1 spk/s; $p=0.69$, Wilcoxon test) or temporal suppression (e.g. 16 vs. 133 msec duration; 78.1±0.8 vs. 81.9±0.8 spk/s; $p<0.001$, Wilcoxon test). We demonstrate that these

pronounced spatiotemporal asymmetries between darks and lights are present over a wide range of luminance conditions (0.1 to 239 cd/m^2) and only disappear under very low mesopic luminance (e.g. 7 vs. 23 deg dark targets at <0.1 cd/m^2; 8.1±1.2 vs. 18.7±0.9 spk/s; $p<0.001$, Wilcoxon test). Our results suggest that ON and OFF visual pathways are specialized in processing different spatiotemporal properties of visual scenes (ON for slow-global change and OFF for fast-local change), a finding that could explain why reflexes triggered by scene movement are ON dominated in many animals (e.g. optokinetic reflex for image stabilization).

Disclosures: **R. Mazade:** None. **J. Jin:** None. **C. Pons:** None. **J. Alonso:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.04/HH33

Topic: D.07. Vision

Support: NIH Grant EY05253

Title: Optical blur affects differently on and off visual pathways

Authors: *C. PONS, R. MAZADE, J. JIN, J. ALONSO
Biol. Sci., State Univ. of New York, New York, NY

Abstract: The eye focuses objects at the plane of fixation leaving large surrounding regions of the scene optically blurred. Image regions that are optically blurred are dominated by low spatial frequencies, which are thought to be less effective at driving ON than OFF visual pathways (Kremkow et al., 2016), however, the effect of optical blur on ON-OFF response balance has not been experimentally studied. Here we measured how ON and OFF responses are affected by optical blur in cat visual cortex, by inducing blur with either contact lenses (-10, 0, +10 diopters) or spectacles (-10, -5, -3, 0, +3, +5, +10 diopters), while using an artificial pupil of 3 mm in all conditions. To quantify the effect of optical blur, we measured changes in receptive field size mapped with light and dark spots and the strength of responses to these stimuli. Our results indicate that the cortical effects of optical blur depend both on the contrast polarity of the stimulus (dark or light) and the polarity of the optical blur (positive or negative). When tested with contact lenses, positive blur made visual responses stronger and cortical receptive fields larger. However, while the response enhancement could be relatively similar for darks and lights (darks: 26.5%, $p<0.0001$, lights: 20.65%, $p<0.0001$, Wilcoxon tests, -10 to +10 diopter), the receptive field enlargement was more pronounced for lights (lights: 17.83%, $p<0.0001$, darks: -0.02%, $p=0.85$, lights-darks: 17.85%, $p<0.0001$, Wilcoxon tests, 0 to +10 diopter). When tested with spectacles, optical blur also made visual responses stronger. However, the effect was dependent on receptive field size, as it would be expected from changes in image magnification

that result from positive (enlargement) and negative (shrinkage) optical power. Spectacles inducing positive blur enhanced the responses of neurons with receptive fields larger than the stimulus while spectacles inducing negative blur enhanced the responses of neurons with smaller receptive fields, and both response enhancements reached saturation at focus. Our results indicate that positive blur (and the associated image magnification) strengthen both OFF and ON visual responses but enlarge more ON than OFF receptive fields. The larger spatial distortion of ON receptive fields could explain why cortical retinotopy is more precise for OFF responses (Kremkow et al., 2016, Li et al., 2016) and why myopia progression is associated with ON visual deficits (Pardue et al., 2008). The saturation of the blur/response function at focus could also provide a mechanism to finely adjust eye optical power to maximally stimulate the smallest receptive fields available in the retina of different animals.

Disclosures: C. Pons: None. **R. Mazade:** None. **J. Jin:** None. **J. Alonso:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.05/HH34

Topic: D.07. Vision

Title: 100 thalamic afferents per cortical point are sufficient to accurately map on and off retinotopy in cat visual cortex

Authors: *E. ZABEH[1], J. JIN[2], R. LASHGARI[1], J. ALONSO[2]
[1]Brain Engin. Res. Ctr., IPM, Tehran, Iran, Islamic Republic of; [2]SUNY Col. of Optometry, New York, NY

Abstract: The thalamocortical pathway is the main route of sensory information to the cerebral cortex, however, we still do not have accurate estimates of how many thalamic afferents converge at each cortical point. Even in cat visual cortex (the animal model with most detailed thalamocortical quantification), the estimates can range from 1000 (Peters and Payne, 1993) to less than 10 (Ringach, 2004; Paik and Ringach, 2011). To provide more realistic estimates of thalamocortical convergence, here we calculated the number of afferents needed to accurately represent ON-OFF retinotopy within a 1-2 mm horizontal track of cat visual cortex. ON-OFF retinotopy was first measured with multielectrode arrays and stimulus spike-trigger-averaging (Neuronexus, 32 electrodes separated by 0.1 mm) and then simulated with a weighted sum of thalamic receptive fields. The simulation used initial random values for receptive field position/size/polarity, and axonal synaptic-weight/cortical-spread that were constrained within physiological ranges (e.g. matched sizes of thalamic/cortical receptive field subregions, average thalamic receptive field scatter per cortical point: 2.5 receptive field centers, equal number of ON and OFF afferents, Jin et al., 2011). After randomizing the initial values, thalamocortical

connections were pruned using a joint-sparse-regression algorithm (Deng et al., 2013) that minimized afferent number without compromising simulation accuracy. The relation between the initial afferent number and simulation accuracy followed a power law that reached r^2: 0.9 with < 100 afferents (average correlation between simulation and data for 80 initial afferents pruned to ~ 65: r^2= 0.98 for OFF, r^2=0.97 for ON, r^2=0.90 for ON-OFF, 10 simulations). When using <100 initial afferents, the simulation accuracy was higher if both excitatory and inhibitory thalamocortical connections were allowed rather than forcing all connections to be excitatory (e.g. r^2 = 0.9 vs. 0.77, for 60 initial afferents). This difference could be reduced but was still maintained when more afferents were used (e.g. r^2 = 0.94 vs. 0.92, for 400 afferents pruned to ~ 125). Reaching a simulation accuracy of r^2 > 0.9 required ~ 40% inhibitory connections for <100 initial afferents (e.g. 41% ON, 40% OFF for 80 afferents) but the percentage was reduced to ~ 34% when the afferent number increased (e.g. 31% OFF, 36% ON, for 400 afferents pruned to ~ 125). We conclude that ON and OFF retinotopy can be accurately mapped in cat visual cortex with a developmental process that uses an excess of thalamic afferents at initial stages, which is pruned to ~100 afferents per cortical point with 30-40% of inhibitory connections.

Disclosures: **E. Zabeh:** None. **J. Jin:** None. **R. Lashgari:** None. **J. Alonso:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.06/HH35

Topic: D.07. Vision

Support: NIH Grant EB022915

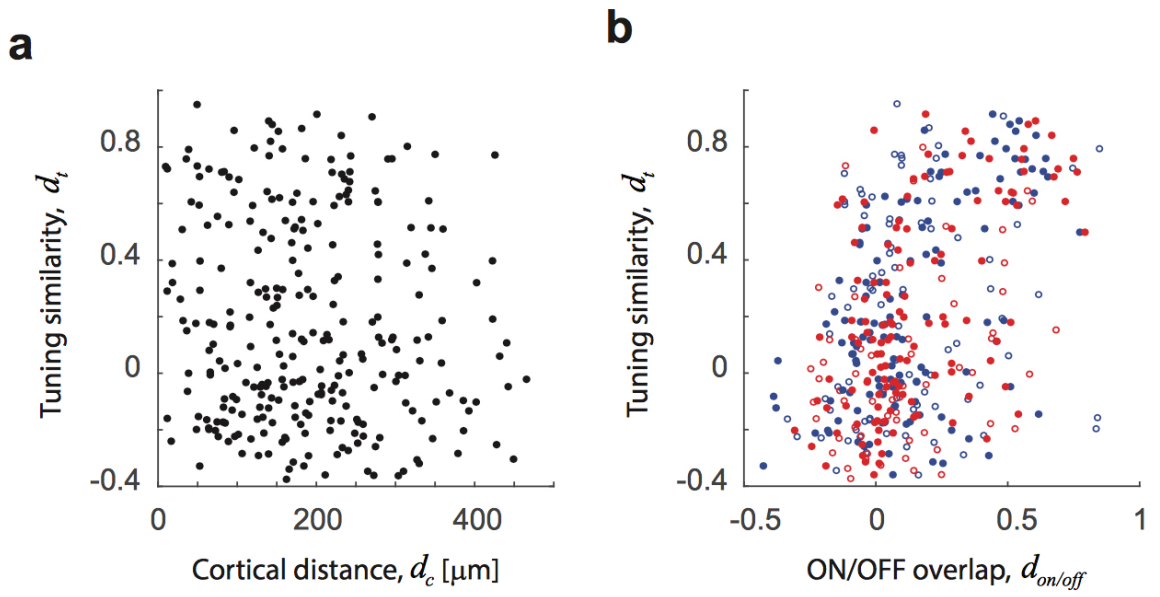
NIH Grant EY018322

NIH Grant EY023871

Title: Untangling cortical maps in mouse primary visual cortex

Authors: *D. L. RINGACH[1], L. JIMENEZ[2], E. TRING[3], J. T. TRACHTENBERG[4]
[1]Neurobio. & Psychology, [2]Psychology, [3]Neurobio., [4]UCLA, Los Angeles, CA

Abstract: Local populations of neurons in mouse primary visual cortex have receptive fields with varied tuning preferences. We show this seeming disorder can be untangled: the similarity of tuning between neurons is correlated better with the overlap of their receptive fields in space rather than with their distance in the cortex. Our findings are consistent with the hypothesis that salt-and-pepper maps in mouse V1 arise in part due to the lateral dispersion of clonally related neurons (Ohtsuki et al, Neuron, 2012).



Disclosures: **D.L. Ringach:** None. **L. Jimenez:** None. **E. Tring:** None. **J.T. Trachtenberg:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.07/HH36

Topic: D.07. Vision

Support: KAKENHI Grant-in-Aid for Young Scientists B 17K14941

Title: 2-P imaging of visual cortex layer 6 corticothalamic feedback in the behaving mouse

Authors: ***S. AUGUSTINAITE**, B. KUHN
Okinawa Inst. of Sci. and Technol. Grad. Univ., Okinawa, Japan

Abstract: Layer 6 (L6), the deepest lamina of cerebral cortex, is one of the key structures regulating behavior state related information processing within the cortex and various subcortical areas. However, very little is known about the functional significance of different L6 circuits in vivo. L6 experiments in the behaving animals still remain challenging due to hard access of the recording / imaging site, complexity of neuronal circuits and heterogeneity in neuron morphology. Here, we focus on primary visual cortex L6 feedback projections to visual thalamus (lateral geniculate nucleus) which regulate visual signal transmission from retina to cortex. We developed a method to study this corticothalamic feedback in vivo with 2P microscopy. With

this method, we can reliably image retrogradely labeled corticothalamic and other excitatory L6 neurons in a head-fixed mouse. Up to a few hundred individual neurons can be recorded simultaneously for several hours and/or repeatedly recorded during different days, while monitoring mouse behavior state. This allows us to study the role of corticothalamic feedback in visual signal processing during different behavior states, ranging from full alertness to sleep.

Disclosures: S. Augustinaite: None. **B. Kuhn:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.08/II1

Topic: D.07. Vision

Title: Distinct patterns of stimulus selectivity, spontaneous activity, and connectivity of L6 callosal projection and thalamic projection neurons in primary visual cortex of the awake mouse

Authors: *Y. LIANG, W. SUN, R. LU, N. JI
Janelia Res. Campus, HHMI, Ashburn, VA

Abstract: The corpus callosum is the major commissural pathway connecting the two cerebral hemispheres in the mammalian brain. In contrast to other cortical regions, within mouse primary visual cortex (V1), callosal connections are dominated by infragranular neurons. Little is known about the properties of these callosal projection neurons or their relationship with the corticothalamic (CT) neurons in layer 6 (L6). Developing an understanding of the physiological roles of these neurons requires dissection of their functional specificity and connectivity. By combining retrograde labeling, in vivo two-photon calcium imaging, and rabies virus-based input mapping, we determined the sensory response properties and upstream monosynaptic connectivity of L6 neurons mediating the callosal cortico-cortical (CC) pathway in V1 of the awake mouse. Distinct from the L6 CT neurons, a smaller percentage of CC neurons are orientation selective (OS), with the OS CC neurons possessing similar orientation tuning properties to those of OS CT neurons. Interestingly, a small fraction of callosal CC neurons, but not CT neurons, show spontaneous activity that is suppressed by the presentation of visual stimuli. Rabies-virus-assisted monosynaptic tracing indicates that callosal CC neurons receive more long-range cortical inputs than CT neurons, consistent with the observation that CC neurons have less confined receptive field than CT neurons. Providing evidence that there is a distinct segregation in the function and connectivity between callosal CC and CT L6 neurons, our results support the notion that L6 is more implicated in corticocortical pathways than what was thought previously.

Disclosures: Y. Liang: None. **W. Sun:** None. **R. Lu:** None. **N. Ji:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.09/II2

Topic: D.07. Vision

Support: SFB870

Title: Integrated circuit analysis of layer 2/3 pyramidal cells in mouse visual cortex

Authors: *S. WEILER, T. ROSE, M. HÜBENER, T. BONHOEFFER, V. SCHEUSS
Dept. Synapses-Circuits-Plasticity, Max Planck Inst. Neurobio., Martinsried, Germany

Abstract: Neocortical pyramidal cells (PCs) display functional specializations defined by their connectivity as well as their molecular, anatomical and electrophysiological properties. PCs in layers 5 (L5) and 6 (L6) have been classified into different subtypes based on their *in vivo* functional response properties and corresponding connectivity patterns as well as genetic and electrophysiological characteristics (Velez-Fort et al. *Neuron* 2014, Kim et al. *Neuron* 2015). For layer 2/3 (L2/3) PCs no unique genetic markers corresponding to individual functional subtypes have been discovered so far (e.g. Zariwala et al. *Front Syst Neurosci* 2010). However, L2/3 PC have been shown to project functionally target-specific to higher visual areas (Glickfeld et al. *Nat Neurosci* 2013).
Here, we ask whether L2/3 PCs differ in their connectivity patterns in mouse primary visual cortex (V1) and whether this is related to differences in L2/3 PC stimulus preferences. To address this question, we characterize the excitatory and inhibitory cortical inputs of the same L2/3 PCs using laser-scanning photostimulation (LSPS) by UV glutamate uncaging in brain slices. The majority of L2/3 pyramidal cells receive strong excitatory input from layer 4 (L4) (77/106 PCs, exc. L4 input >50% of total input) and strong local inhibitory input from L2/3 (67/106 PCs, inh. L2/3 input >50% of total input) and varying degrees of additional input from the other layers (e.g. 19/106 PCs, exc. L5 input >25% of total input). Furthermore, the spatial overlap between excitatory and inhibitory synaptic input within a given L2/3 PC varies, e.g. L5 excitatory input is mostly unbalanced by L5 inhibitory input.
In order to explore the functional implications of the different input patterns we developed an *in vivo* / *in vitro* approach: First, we characterize the visual response properties (orientation/direction selectivity, temporal/spatial preferences, ocular dominance and spontaneous activity) of individual neurons expressing genetically encoded calcium indicators (GECIs) with *in vivo* 2-photon calcium imaging. Subsequently, we retrieve the same neurons in brain slices for circuit analysis with LSPS.
In conclusion, L2/3 PCs appear to be a heterogeneous group based on their connectivity patterns, and we currently explore if this is reflected in their visual stimulus preferences.

Disclosures: **S. Weiler:** None. **T. Rose:** None. **M. Hübener:** None. **T. Bonhoeffer:** None. **V. Scheuss:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.10/II3

Topic: D.07. Vision

Title: Neocortical layer 5 is composed of cell type specific microcolumnar circuits

Authors: *S. SAKAI[1,2], H. MARUOKA[2], N. NAKAGAWA[2], S. TSURUNO[2], T. YONEDA[2], T. HOSOYA[2]
[1]Tokyo Metropolitan Inst. of Med. Sci., Tokyo, Japan; [2]Lab. for Local Neuronal Circuits, RIKEN BSI, Wako, Saitama, Japan

Abstract: Neocortex contains various types of excitatory and inhibitory neurons, each with specific axonal projection patterns, gene expression, cell morphology and electrophysiological properties. Understanding the structure of microcircuits composed of these neuronal types is a key to investigate neocortical information processing. Layer 5, an output layer of neocortex, contains two major types of projection neurons. Subcerebral projection neurons (SCPNs) project axons to extracortical targets including the pons, superior colliculus and spinal cord, while cortical projection neurons (CPNs) project axons to other cortical areas. We have previously reported that SCPNs form periodic columnar clusters termed microcolumns and that SCPNs in the same microcolumns show similar visual responses (Maruoka et al., J Neurosci 2011). These observations suggest that SCPN microcolumns function as functional units in layer 5. It has been unknown, however, whether CPNs and inhibitory neurons in layer 5 form microcolumnar functional units similarly to SCPNs. We conducted three-dimensional analysis of the spatial arrangement of major neuronal types in layer 5. Each neuron type was specifically labeled using retrograde tracers, transgenic mice expressing marker proteins or immunostaining. After brain samples were fixed and cleared, large areas of the cortex were imaged by two-photon microscopy. Both SCPNs and CPNs showed thin columnar arrangement in a wide range of cortical areas. These SCPN and CPN microcolumns were interdigitated with each other. We performed in vivo calcium imaging to investigate the activity of the cell type specific microcolumns. Columnar correlations of spontaneous activity and visual response preference were analyzed. Our result indicates that the neocortical layer 5 is composed of cell type specific microcolumnar circuits.

Disclosures: **S. Sakai:** None. **H. Maruoka:** None. **N. Nakagawa:** None. **S. Tsuruno:** None. **T. Yoneda:** None. **T. Hosoya:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.11/II4

Topic: D.07. Vision

Support: JSPS KAKENHI Grant Number JP16K14565

Title: Involvement of gap junctions in the function of microcolumns in neocortex

Authors: *N. NAKAGAWA, T. YONEDA, H. MARUOKA, S. SAKAI, T. HOSOYA
Lab. for Local Neuronal Circuits, RIKEN Brain Sci. Inst., Wako-Shi, Saitama-Ken, Japan

Abstract: Gap junctions mediate electrical coupling between neurons and promote neuronal circuit formation. Recently we found that subcerebral projection neurons (SCPNs), one of the major types of excitatory neuron in layer 5, selectively couple with radially aligned SCPNs in the first postnatal week. The coupling was cell-type-specific; SCPNs rarely coupled with cortical projection neurons, another major type of excitatory neuron. The coupling disappeared during the next week, suggesting its involvement in the development of neuronal circuits but not in the adult brain functions.In a wide range of cortical areas, SCPNs form radial clusters termed microcolumns with a diameter of 1-2 cells, suggesting that the gap junctions preferentially couple SCPNs in individual microcolumns. Our recent analyses revealed that SCPNs in individual microcolumns exhibit correlated activity in the adult stage in vivo. These results raise the possibility that the gap junction network is involved in the construction of microcolumnar neuronal circuits that promote correlated activity in adults.To test this hypothesis, we conducted forced expression of mutant connexin (DNCx) that suppresses electrical coupling in a dominant-negative manner. DNCx expression partially but significantly suppressed electrical coupling between SCPNs in the early postnatal stage. Our preliminary results suggest that DNCx expression impaired correlated activity in the adult stage without altering the microcolumnar organization or basic neuronal activities. These results suggest that electrical coupling between SCPNs promote the development of microcolumnar circuits that enable correlated activity, and thereby establishing the behavior of microcolumns as functional units in neocortex.

Disclosures: **N. Nakagawa:** None. **T. Yoneda:** None. **H. Maruoka:** None. **S. Sakai:** None. **T. Hosoya:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.12/II5

Topic: D.07. Vision

Support: NIH Grant EY024946

NIH Grant EY018251

Title: LGN input to fast-spike interneurons in layers 4 and 6 of awake rabbit visual cortex

Authors: *Y. I. BERESHPOLOVA[1], X. HEI[1], C. R. STOELZEL[1], J.-M. ALONSO[2,1], H. A. SWADLOW[1,2]
[1]Dept. of Psychology, Univ. of Connecticut, Storrs, CT; [2]Biol. and Visual Sci., SUNY Optometry, New York, NY

Abstract: Fast-spike interneurons in layer 4 (L4) of rabbit visual cortex (V1) receive monosynaptic input from the LGN and convey a fast, sensitive, and broadly tuned feed-forward inhibition onto local targets. Layer 6 (L6) also receives a distinct, albeit weaker input from the LGN, and contains fast-spike interneurons. To understand better the role of these interneurons in the thalamocortical circuit, we examine the rules governing thalamocortical connectivity and sensory response properties of putative fast-spike interneurons (suspected inhibitory interneurons, SINs) in both input layers of awake rabbits. We found that L6 SINs, like those of L4 (Zhuang et al., 2013), were very broadly tuned to stimulus orientation/direction, had complex receptive fields (RF) with extensively overlapping ON and OFF zones, responded in a similar, non-linear manner to drifting visual gratings (F1/F0 ratios < 1), showed similar tuning for spatial/temporal frequency, and had high spontaneous firing rates. However, visual responses of L6 SINs were much less sensitive to visual stimuli (higher C50 values: 25.86 ± 2.95 % vs. 10.83 ± 1.94 %, $p < 0.001$, Wilcoxon rank-sum test), responded less robustly to drifting visual grating (lower F1 values: 11.54 ± 1.56 spikes/s vs. 32.97 ± 3.73 spikes/s, $p < 0.001$, Wilcoxon rank-sum test), and were less reliable (higher Fano factors: 2.18 ± 0.34 vs. 1.33 ± 0.08, $p = 0.025$, Wilcoxon rank-sum test) than those of L4 SINs. Cross-correlation analyses of LGN neurons and L4/L6 SINs showed a high connection probability that was strongly related to the degree of retinotopic overlap, with little dependence on matching either (a) the preferred sign of the LGN cell (on-center or off-center) and the SIN (on-dominated or off-dominated), or (b) the sustained/transient response classification of the LGN/SIN paired cells. In both rabbits (Swadlow and Gusev, 2002) and rats (Bruno and Simons, 2002), L4 fast-spike interneurons receive a strong, highly convergent/divergent input from neurons in the topographically aligned thalamic region. Our results show that this is also true for fast-spike interneurons in both L4 and L6 of the visual cortex. Although L6 SINs of V1, like those in L4, are well-suited to convey a broadly

tuned feed-forward inhibition onto their local targets, their responses to visual stimuli are considerably less sensitive, less reliable, and less responsive to changing stimulus conditions.

Disclosures: Y.I. Bereshpolova: None. **X. Hei:** None. **C.R. Stoelzel:** None. **J. Alonso:** None. **H.A. Swadlow:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.13/II6

Topic: D.07. Vision

Support: Wellcome Trust 095668

Wellcome Trust 095669

Marie Curie IEF 627787

Title: A circuit for spatial integration and its modulation by running in four classes of V1 neurons

Authors: *M. DIPOPPA, M. CARANDINI, K. D. HARRIS
Univ. Col. London, London, United Kingdom

Abstract: A prominent feature of neurons in primary visual cortex (V1) is selectivity for the size of visual stimuli, known as size tuning. In pyramidal (Pyr) neurons of mouse V1, size tuning is thought to be imposed by somatostatin-expressing (Sst) interneurons, whose responses integrate Pyr input over a wide cortical area. Size tuning, moreover, depends on locomotion, which can profoundly affect sensory responses in multiple neuronal classes of V1. We sought to understand the interplay between these phenomena by measuring the activity of Pyr, Sst, Parvalbumin (Pvalb), and Vasoactive Intestinal Polypeptide (Vip) cells, and describing their interactions with a simple circuit model.
We imaged the visual responses of superficial V1 neurons with the calcium indicator GCaMP6 using a 2-photon microscope, identifying Pyr, Sst, Vip and Pvalb neurons in transgenic mice. We measured size tuning both for cells whose receptive field was centered on the visual stimulus and for the remaining off-center cells. In contrast with a previous report, well-centered Sst neurons did exhibit size tuning, similarly to the other cell classes. Only off-center Sst neurons increased monotonically their responses with stimulus size. Additionally, locomotion increased only responses to small stimuli in Vip neurons and only responses to large stimuli in Sst neurons.
To capture these observations we devised a rate model based on the recurrent connections of these neurons classes and on feedforward excitation from thalamus. In the model, the visual responses of each class is estimated as the sum of average responses from local Pyr and thalamic

cells subtracted or divided by the average responses of functionally connected interneuron classes. The inputs are integrated over a cortical area of finite size with two-dimensional Gaussian functions.
The model provided accurate quantitative fits to the data of all cell types, but only if we incorporated the following constraints of the functional connections between V1 cell classes: (1) a broad spatial integration of inputs from Sst neurons to Pyr neurons; (2) thalamic visual input that impinges not only on Pyr and Pvalb neurons but also on Sst cells (perhaps through Pyr neurons in deeper layers); (3) a divisive effect of Sst cells onto Vip cells; (4) a subtractive effect of Vip cells onto Sst cells; (5) thalamic visual input that becomes stronger with locomotion (perhaps due to stronger thalamocortical synapses). We conclude that a relatively simple rate model with reasonable assumptions can capture the apparently complex size- and locomotion-dependent responses of multiple classes of V1 neurons.

Disclosures: **M. Dipoppa:** None. **M. Carandini:** None. **K.D. Harris:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.14/II7

Topic: D.07. Vision

Support: Wellcome Trust 4 years PhD 099692/Z/12/Z

Wellcome Trust 095669

Wellcome Trust 095668

Simons Foundation (SCGB 325512)

Title: Functional organization of presynaptic networks in primary visual cortex

Authors: *L. F. ROSSI, K. D. HARRIS, M. CARANDINI
Univ. Col. London, London, United Kingdom

Abstract: Neurons in the primary visual cortex (V1) are tuned to retinotopic location, orientation and direction of motion. Such selectivity stems from the integration of inputs from hundreds of presynaptic neurons distributed across cortical layers. Yet, the functional principles that organize such presynaptic networks have only begun to be understood. Is the presynaptic network uniform in space, or are there anisotropies, perhaps related to the postsynaptic neuron's orientation preference? Does dendritic morphology contribute to such anisotropies? How does the stimulus preference of a postsynaptic neuron relate to the activity of its presynaptic partners?
To answer these questions, we used monosynaptic rabies virus tracing and two-photon

microscopy in mouse V1. We bred mice expressing the calcium indicator GCaMP6 in all excitatory cortical neurons, and we electroporated a pyramidal neuron in L2/3 (starter neuron) with plasmids coding for the rabies glycoprotein oG, the avian receptor TVA and the fluorescent protein dsRed. Then we extracellularly injected an EnvA-dG-dsRed rabies virus, which could only infect the TVA tagged starter neuron and then spread to its presynaptic partners. Finally, we imaged the activity of the starter cell, its presynaptic neurons and the surrounding excitatory population across layers 2-5 in response to sparse noise or drifting gratings, while the mouse was free to run on a treadmill. We also acquired high resolution z-stacks to reconstruct the morphology of the starter neuron and the position of its inputs.
For each starter neuron, we identified on average 132±41 local V1 presynaptic partners (range of 38-334, n = 8), distributed between L1 and L5 and most abundant in layers 2/3 and 4. The connection probability to neighboring neurons decreased laterally with a space constant of 128±16μm. The resulting distribution of presynaptic spanned 46±3 degrees in azimuth and 26±1 degrees in elevation of the visual field. In comparison, the density of dendrites fell laterally with a space constant of 83±17μm.
Many presynaptic neurons (53±7%) displayed activity-dependent fluorescence changes: 32±2% of these neurons (17±3% of the total) responded significantly to drifting gratings, as did the majority of the starter neurons (6/8). Our preliminary analysis shows that the tuning of neurons in each presynaptic network is heterogeneous and is not necessarily aligned to the orientation preference of the corresponding starter neuron.
We are currently investigating further the relationship between visual properties of the starter neurons and the functional organization of their presynaptic networks.

Disclosures: **L.F. Rossi:** None. **K.D. Harris:** None. **M. Carandini:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.15/II8

Topic: D.07. Vision

Support: HFSP LT000173/2013

SNF Ambizione

Title: Single-cell-initiated monosynaptic tracing reveals diverse network properties in visual cortex

Authors: ***S. TRENHOLM**[1,2], A. WERTZ[2], A. BHARIOKE[2], K. YONEHARA[3], Z. RAICS[2], D. HILLIER[2], J. JÜTTNER[2], M. LEINWEBER[2], A. OLIVEIRA[3], G. SZALAY[4], G. KELLER[2], B. RÓZSA[4], B. ROSKA[2,5]

[1]McGill Univ., Montreal, QC, Canada; [2]Friedrich Miescher Inst., Basel, Switzerland; [3]Danish Res. Inst. of Translational Res., Aarhus, Denmark; [4]Hungarian Acad. of Sci., Budapest, Hungary; [5]Univ. of Basel, Basel, Switzerland

Abstract: Introduction: Individual neurons in cortex receive inputs from hundreds of neurons. How the response properties of a single cortical neuron relates to those of its presynaptic partners remains poorly understood. **Methods:** We performed single-cell-initiated, monosynaptically-restricted, retrograde transsynaptic tracing with modified rabies viruses expressing GCaMP6s to trace the presynaptic networks of individual neurons in layer 4 of mouse visual cortex. We then performed in vivo calcium imaging of the postsynaptic neuron and of its presynaptic network during visual stimulation. **Results:** This technique labelled hundreds of presynaptic cells connected to individual layer 4 neurons in primary visual cortex, with the majority of presynaptic neurons being located nearby in primary visual cortex. Using in vivo calcium imaging, we characterized the response properties of the starter neuron and its presynaptic network to a diverse set of visual stimuli. We found that presynaptic networks connected to individual layer 4 neurons could be classified into different functional groups based on their response properties. In addition, we compared the functional response properties and anatomical locations of presynaptic neurons belonging to individual layer 4 neurons with previous data regarding individual layer 2/3 postsynaptic neurons. **Conclusion:** These results reveal the existence of diverse network organization principles belonging to the presynaptic networks of individual layer 4 pyramidal neurons in mouse visual cortex.

Disclosures: S. Trenholm: None. **A. Wertz:** None. **A. Bharioke:** None. **K. Yonehara:** None. **Z. Raics:** None. **D. Hillier:** None. **J. Jüttner:** None. **M. Leinweber:** None. **A. Oliveira:** None. **G. Szalay:** None. **G. Keller:** None. **B. Rózsa:** F. Consulting Fees (e.g., advisory boards); Femtonics (scientific advisory board). Other; Author founded a company (Femtonics) that produces a product or service related to the work being reported. **B. Roska:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.16/II9

Topic: D.07. Vision

Support: Strategic Priority Research Program, XDB02020001

Strategic Priority Research Program, XDB02040100

Natural Science Foundation of China, 30921064, 90820008, and 31130027

Shanghai Key Basic Research Project (16JC1420201)

973 Program, 2011CBA00400

Title: Functional organization of intrinsic and feedback presynaptic inputs in the primary visual cortex

Authors: *Y. WEN, Q.-F. ZHANG, H. LI, A. GUO, M.-M. POO
Inst. of Neuroscience, SIBS, CAS, Shanghai City, China

Abstract: In the primary visual cortex (V1) of mammals with highly developed visual systems, neurons are often organized according to their preferred stimulus orientation into a highly ordered map. However, whether and how the various presynaptic inputs to V1 neurons are organized relative to the neuronal orientation map remains unclear. To address this issue, we constructed genetically encoded calcium indicators targeting axon boutons, and used them to directly map the organization of axon boutons of V1 intrinsic and V2-V1 feedback projections in adult tree shrews. Both connections are spatially organized into maps according to the preferred orientations of the axon boutons. Dual-color calcium imaging showed that V1 intrinsic inputs are more precisely aligned to the orientation map of V1 cell bodies than V2 feedback projections. Such spatially organized axon bouton maps could provide substantially clustered inputs to dendrites of V1 neurons and facilitate the establishment of V1 cell body map.

Disclosures: Y. Wen: None. **Q. Zhang:** None. **H. Li:** None. **A. Guo:** None. **M. Poo:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.17/II10

Topic: D.07. Vision

Support: National Eye Institute Grant RO1-EY-109272

Simons Foundation

March of Dimes Foundation

Title: Local interneurons form an organizing nidus for feature-selective subnetworks in mouse visual cortex

Authors: *G. S. PALAGINA[1], S. M. SMIRNAKIS[2]
[1]Neurol., Boston VA Res. Inst., Boston, MA; [2]Brigham and Women's Hospital, Harvard Med. Sch., Boston, MA

Abstract: Sensory stimuli are encoded by the joint firing of neuronal groups rather than single isolated neurons. However, the rules by which these groups encode information remain poorly

understood. A leading hypothesis is that similarly tuned interconnected pyramidal cells form multicellular encoding units yoked to a similar purpose. The existence of such units likely reflects on the profile of the spontaneous population bursts in neocortical networks. We studied the population activity of pyramidal cells and interneurons in layer 2/3 of the mouse area V1 over the course of visual cortical maturation (postnatal days 7-45). We recorded spontaneous events and visually evoked neuronal responses over several hours using two-photon calcium imaging. The sizes of spontaneous population bursts in layer 2/3 formed scale-free distributions obeying a power law. This was also true for the degree of functional connectivity (a measure of pairwise synchrony) across layer 2/3 neurons. These findings are consistent with the hierarchical small-world network architecture, where cliques of highly interconnected cells ("small worlds") communicate with each other via a restricted number of links between "hub" cells. To detect candidate "small world" cliques we focused on the groups of pyramidal cells that: (a) had significant functional connectivity between each other (displayed high synchrony), and, (b) generated calcium events that showed a consistent temporal relationship with the events of specific local interneurons in the context of ongoing spontaneous activity. This approach was guided by the intuition that neurons whose synchronous firing represents a "conceptual unit" ought to be inhibited together. We thus identified cliques of pyramidal neurons temporally "linked" to one or more local interneurons. These cliques were already present before eye opening, but did not remain static during postnatal development: both clique size and member overlap with other cliques decreased over time, as pyramidal neurons became progressively more selective, "linking" to fewer local interneurons. Notably, pyramidal neurons in a clique shared functional properties (tuning for orientation and direction of motion) between each other and with their partner interneuron. Our findings suggest that spontaneous population bursts in the V1 are shaped by "small-world" networks of pyramidal neurons that share functional properties and work in concert with one or more local interneurons. We propose that such groups represent a fundamental neocortical unit of computation at the population level.

Disclosures: G.S. Palagina: None. **S.M. Smirnakis:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.18/II11

Topic: D.07. Vision

Support: MINECO-ERDF Grant BFU2014-58776-r

MINECO Grant SEV-2013-0317

Title: Thalamic influence on the statistical wiring of visual cortical receptive fields and maps

Authors: A. J. VALIÑO[1], J. R. BROTONS-MAS[1], F. T. SOMMER[2], J. A. HIRSCH[3], S. SALA[1], *L. M. MARTINEZ[1]
[1]Inst. De Neurociencias De Alicante. CSIC-UMH, Sant Joan d'Alacant, Spain; [2]Univ. California, Helen Wills Neurosci Inst., Helen Wills Neurosci. Inst., Berkeley, CA; [3]USC, Los Angeles, CA

Abstract: Neurons in the primary visual cortex (V1) of most mammals respond only to a restricted set of stimulus orientations. Furthermore, in some species but not in others, V1 cells with similar orientation preferences tend to cluster together in an orderly fashion giving rise to the renown cortical orientation preference maps (OPMs). Over the years, contrasting models emphasizing the role of feedforward vs. intracortical connectivity have been proposed to explain the emergence and function of this salient feature of cortical organization. Still highly debated, both types of developmental models have, nonetheless, in common that they largely neglect the potential role that the thalamus (lateral geniculate nucleus, LGN) plays in this process. Recently, we have demonstrated that the retinothalamic circuit is optimized to increase the resolution of the retinal output on its way to V1 through a straight process of information upsampling and interpolation occurring at the level of the LGN (Martinez et al., 2014). Here, we use a similar approach to show that the probabilistic, convergent connectivity from retina to LGN required to increase visual resolution significantly transforms the thalamic representation of the retinal mosaics generating partially segregated thalamic domains of On- and Off-center receptive fields cells arranged in locally correlated clusters of dipoles with different orientations. We further demonstrate that this new thalamic structure has the same properties as those previously shown in cortical orientation maps and suggest that it might be essential for their emergence and stability: First, our results revealed that the structure of the emergent cortical maps perfectly correlates very well with the arranging of the thalamic ON and OFF domains. Second, the periodicity and stability of the cortical orientation map depend critically on the biological constraint imposed by the upsampling and interpolation procedure performed in the LGN. Finally, the retinothalamic rewiring allows to maintain large values of thalamocortical convergence, and still conserving the stability of the map, without requiring complex developmental rules or very precise patterns of spontaneous or visually driven feedforward activity.

Disclosures: A.J. Valiño: None. **J.R. Brotons-Mas:** None. **F.T. Sommer:** None. **J.A. Hirsch:** None. **S. Sala:** None. **L.M. Martinez:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.19/II12

Topic: D.07. Vision

Title: Signatures of colorvision in the functional architecture of the primary visual cortex

Authors: *M. SCHOTTDORF, J. LIEDTKE, F. WOLF
MPI DS, Goettingen, Germany

Abstract: Color vision was lost in mammals during the nocturnal bottleneck when our ancestors were small, dark-dwelling animals between 205 to 65 Million years ago (Ma). Among modern mammals old world monkeys and great apes (re-)invented trichromacy 30-40 Ma. The newly developed color vision inserted new pathways into cortical functional architecture, potentially perturbing the layout of orientation domains in the primary visual cortex (V1) through non-orientation selective cytochrome oxidase blobs. How much impact color vision had on the overall functional architecture of V1 remains unclear because most studies are phenomenological and little is known about theoretically expected effects. Here we assess the experimental signatures of two distinct and paradigmatic model types to fill in this gap.
The first one is a coordinated optimization model of visual cortical maps, in which the coupling between the layouts of orientation and color are expanded (Reichl et al. PLoS CB 2015, Bressloff et al. PRL 2002). We find that this type of interaction leaves a strong signature in the pinwheel configuration, detectable with the measures of the common design (Kaschube et al. Science 2010, Schottdorf et al. PLoS CB 2015). The second model is a phenomenological model to incorporate non-orientation selective regions into a layout of orientation domains by geometric distortions. Models of this type leave the measures of the common design invariant, but generate higher order layout correlations.
In summary, we discovered a set of quantitative, specific and measurable predictions from both models that can in principle be falsified given the precision of available data. By defining these most informative measures we provide a solid foundation for a future comparative studies across mono-, di- and trichromatic mammals that will help to clarify whether there is any signature of colorvision in the functional architecture of V1.

Disclosures: **M. Schottdorf:** None. **J. Liedtke:** None. **F. Wolf:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.20/II13

Topic: D.07. Vision

Support: NIH NS093998

Title: Mapping visual cortical processing networks with infrared neural stimulation

Authors: *M. M. CHERNOV[1], R. M. FRIEDMAN[2], A. W. ROE[3,4]
[1]Neuroscience, Oregon Natl. Primate Resource Ctr., Oregon Hlth. and Sci. Univ., Beaverton, OR; [2]Div. of Neurosci., Oregon Hlth. & Sci. Univ. - ONPRC, Beaverton, OR; [3]Zhejiang Univ., Zhejiang, China; [4]Neurosci., Oregon Hlth. and Sci. University- ONPRC, Beaverton, OR

Abstract: Microelectrodes for brain mapping have been the gold standard for over a century. However, the technique suffers from several drawbacks, such as poor spatial precision and technical issues that arise when this type of stimulation is combined with state of the art recording methods, such as electrophysiology or MRI. Over the past two decades, significant progress has been made into optical alternatives to electrophysiological techniques. One such method is infrared neural stimulation (INS), which uses short (on the order of 10^{-4} s) pulses of 1875 nm light to create focal thermal transients that lead to generation of action potentials in both the CNS and peripheral nerves (Wells et al. 2005, Opt. Lett., Cayce et al. 2014, Neuroimage, Chernov and Roe 2014, Neurophotonics). Because light at this wavelength is quickly absorbed by water in tissue, the stimulation is essentially confined to the radius of the illuminating spot produced by light from an optical fiber and spreads a few hundred microns into tissue. The high spatial precision of the method, its minimally invasive nature (stimulation can be contact-free) and its compatibility with a variety of imaging modalities including intrinsic optical imaging (IOS) and MRI, as well as electrophysiological recordings make it an excellent tool for the study of functional connections in cortex.
We are interested in using INS as a probe to reveal the organization of functional connections in primate sensory cortices when used in combination with IOS and other imaging methods. Previously, we have found that ocular dominance (OD) eye columns in visual cortex can show enhanced activation following INS (Cayce et al. 2011, Neuroimage). In this study, we use focal (~200-micron diameter illumination spot) INS to reveal short-range projections between functional domains in primary visual cortex, including OD columns, orientation domains and blobs when applied combination with relevant visual stimuli. We report that focal INS leads to patches of activation that line up with the location of functional domains. Moreover, we show that the strength of these connections is modulated by visual input. We conclude that INS is a useful tool for *in vivo* functional dissection of cortical circuits.

Disclosures: M.M. Chernov: None. **R.M. Friedman:** None. **A.W. Roe:** None.

Poster

402. Visual Cortex: Functional Architecture and Circuits

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 402.21/II14

Topic: D.07. Vision

Title: Organization of orientation and spatial frequency preferences in V1: Two-photon imaging of awake monkey

Authors: *Y. SHAO[1,2], S.-C. GUAN[3], N.-S. JU[2], L. TAO[2], C. YU[3,4,5], S.-M. TANG[2,4]
[1]Peking Univ., Beijing City, China; [2]Sch. of Life Sci., [3]Peking-Tsinghua Ctr. for Life Sci., [4]McGovern Inst. for Brain Res., [5]Dept. of Psychology, Peking Univ., Beijing, China

Abstract: Both intrinsic and fluorescence optical imaging have revealed highly organized functional maps of orientation and spatial frequency preference in cat and monkey V1. Here we use two-photon calcium (GCaMP5) imaging of awake, fixating monkey to investigate the response properties of layers 2 and 3 (depths of 150 um and 300 um) V1 neurons. The visual stimulus consists of Gabor gratings at 12 orientations (-45-120 deg), 6 spatial frequencies (0.25-8 cpd) at 90% contrast and drifting at 2 cycles/sec. We recorded from 1475 layer-2 and 1194 layer-3 neurons (orientation OSI > 0.5, ANOVA $p < 0.05$) at two sites (both within a window of 850x850 um at 3-deg parafovea). We found that orientation and spatial frequency maps are largely orthogonal at both sites and both depths, confirming previous results. We also found a systematic decrease of both orientation and spatial frequency tuning bandwidths (half-width at half maximum) as we move away from pinwheel centers. Both layers 2 and 3 contain highly orientation selective neurons, with roughly 9% of the neurons with orientation bandwidths less than 15 degrees and 31% of the neurons with bandwidths less than 30 degrees. Both layers 2 and 3 contain mainly medium frequency neurons (with optimal frequencies between 1-4 cpd), which make up roughly 80% of both layers. However, we observed a significant peak spatial frequency response shift to lower spatial frequencies as we move from layer 3 to 2, with only 5% of layer 3 neurons but over 9% of the layer 2 neurons peaking at less than 1 cpd. Thus while orientation selectivity remain largely invariant as we move from layer 3 to 2, spatial frequency tuning moves to lower spatial frequencies, indicating the role of the laminar V1 structure in shaping lower spatial frequency processing.

Disclosures: Y. Shao: None. **S. Guan:** None. **N. Ju:** None. **L. Tao:** None. **C. Yu:** None. **S. Tang:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.01/II15

Topic: D.07. Vision

Support: ANR-13-JSV4-0007-01

ANR-12-BSV4-0005

Title: A new visuotopic cluster in macaque posterior parietal cortex revealed by wide-field retinotopy

Authors: *S. RIMA, B. R. COTTEREAU, Y. HÉJJA-BRICHARD, Y. TROTTER, J.-B. DURAND
Cerco, Toulouse, France

Abstract: The visuotopic organization of macaque posterior parietal cortex (PPC) is investigated with functional imaging (fMRI) and wide-field retinotopic mapping. A new cluster of 4 visuotopic areas is revealed at the confluence of the parieto-occipital and intra-parietal sulci, in a location that has previously been defined histologically and anatomically as the posterior intra-parietal (PIP) region. This PIP cluster groups together 2 newly identified areas (PIP1/2) and 2 recently described areas (CIP1/2). it is bordered by other visuotopic areas: V3A/DP laterally, V6/V6A medially and LIP anteriorly. Together, these results show that monkey PPC is endowed with a dense set of visotopic areas, as its human counterpart. The fact that these areas can be robustly mapped with fMRI and wide-field stimulation offers both a new framework for invasive investigations in monkey PPC and a promising avenue for direct comparisons with human PPC.

Disclosures: S. Rima: None. **B.R. Cottereau:** None. **Y. Héjja-Brichard:** None. **Y. Trotter:** None. **J. Durand:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.02/II16

Topic: D.07. Vision

Support: McGovern Institute for Brain Research

Title: Functional parcellation of lateral prefrontal cortex in macaques

Authors: *R. RAJIMEHR[1], H. XU[1], D. Y. TSAO[2], R. DESIMONE[1]
[1]MIT, Cambridge, MA; [2]Caltech, Pasadena, CA

Abstract: Prefrontal cortex in primates plays a crucial role in high-level cognitive processes such as working memory, rule-based learning, abstract categorization, attentional control, and decision-making. Prefrontal cortex has been divided into distinct regions based on cytoarchitectonics and connectivity patterns. The visually-responsive regions are located in lateral prefrontal cortex. Here we used a data-driven approach to functionally parcellate lateral prefrontal cortex in macaque monkeys based on fMRI responses to a naturalistic movie stimulus. In this approach, all vertices/points of lateral prefrontal cortex were first represented in a high-dimensional activity space. Vertices were then projected into a low-dimensional space using a

combination of principal and independent component analyses. Hierarchical clustering of vertices revealed spatially distinct clusters that corresponded to areas FEF+45, 46, and dorsal prefrontal. Clustering analysis also showed patches in premotor cortex, including a patch in ventral premotor cortex that appeared to be the 'mirror-neuron' region. The ventral premotor cluster showed higher response to body movements and action scenes in the movie, and it was selectively connected to non-frontal regions involved in action representation. Our approach provides a new look at the functional organization of prefrontal cortex during natural vision.

Disclosures: **R. Rajimehr:** None. **H. Xu:** None. **D.Y. Tsao:** None. **R. Desimone:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.03/II17

Topic: D.07. Vision

Support: EU Grants 604102, 720270 (HBP)

Helmholtz portfolio theme SMHB

VSR computation time grant JINB33

DFG SFB 936/A1, Z3; TRR 169/A2

EU Grant 269921 (BrainScaleS)

Jülich Aachen Research Alliance (JARA)

Title: Elucidating multi-scale cortical dynamics via simulation: A multi-area spiking model of macaque visual cortex

Authors: *S. J. VAN ALBADA[1], *S. J. VAN ALBADA[1], M. SCHMIDT[2], R. BAKKER[1,3], K. SHEN[4], G. Y. BEZGIN[5], C. C. HILGETAG[6,7], M. DIESMANN[1,8]
[1]Res. Ctr. Juelich, Juelich, Germany; [2]RIKEN Brain Sci. Inst., Wako, Japan; [3]Radboud Univ., Nijmegen, Netherlands; [4]Rotman Res. Inst., North York, ON, Canada; [5]Montreal Neurolog. Inst., McConnell Brain Imaging Ctr., Montreal, QC, Canada; [6]Univ. Med. Ctr. Hamburg-Eppendorf, Hamburg, Germany; [7]Dept. of Hlth. Sci., Boston Univ., Boston, MA; [8]RWTH Aachen Univ., Aachen, Germany

Abstract: Spontaneous cortical activity is structured at multiple scales, showing phenomena such as oscillations, propagation of activity, resting-state networks, and area-specific intrinsic time scales. Most of these phenomena rely on inter-area interactions. Moreover, patterns of cortical interactions and dynamics are layer- and cell-type-specific. Thus, macroscopic

interactions as well as microscopic cortical organization need to be taken into account for modeling multi-scale cortical dynamics.
We present a spiking model of all vision-related areas in one hemisphere of primate (macaque) cortex. Each area is represented as 1 mm^2 of cortex, with the full density of neurons and synapses, avoiding distortions of the dynamics due to downscaling [1]. Individual neurons are described as integrate-and-fire nodes. The local circuits cover layers 2/3, 4, 5, and 6, each with an excitatory and an inhibitory population. The model, comprising 4 million neurons, is implemented in NEST and simulated on the JUQUEEN supercomputer.
Model construction follows an integrated bottom-up and top-down approach. Axonal tracing data and statistical regularities define the connectivity. A constant density of synapses along with a gradient of decreasing neuron density from eulaminate to limbic areas yields a gradient in indegrees (the numbers of synapses per neuron). Synapses are statistically mapped to the soma locations of target neurons based on morphological reconstructions, leading to non-negligible feedback onto layer 4 neurons. Slight adjustments of the connectivity based on mean-field theory stabilize the network at realistic firing rates [2]. The result is a full population-level connectivity matrix for macaque visual cortex.
Spiking activity of the model fluctuates on multiple time scales. For intermediate cortico-cortical synaptic weights, the network shows metastable activity and its functional connectivity resembles that of empirical fMRI data. Intrinsic time scales are short in primary visual cortex and longer in more limbic areas, akin to experimental observations. Time scales are influenced by both the gradient in indegrees across areas and inter-area interactions. Activity propagates mainly in the feedback direction, similar to non-sensory-driven brain states. Granger causality analysis reveals strong feedback influences from layer 5 to layer 6. The consistency of the model with a wide range of observations renders it a promising basis for further enhancing our understanding of cortical dynamics.
1. van Albada SJ, Helias M, Diesmann M. PLoS CB 2015 11(9):e1004490.
2. Schuecker J, Schmidt M, van Albada SJ, Diesmann M, Helias M. PLoS CB 2017 13(2):e1005179.

Disclosures: **S.J. Van Albada:** None. **M. Schmidt:** None. **R. Bakker:** None. **K. Shen:** None. **G.Y. Bezgin:** None. **C.C. Hilgetag:** None. **M. Diesmann:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.04/II18

Topic: D.07. Vision

Support: FWO G0A5613N

FWO G043912N

IUAP7/11

PFV/10/008

HBP 604102 (EU H2020)

QZ is a postdoctoral fellow of the FWO-Flanders

Title: Dorsal visual cortex in macaques resembles that of New World monkeys: A reassessment of its topography based on sub-millimeter retinotopic fMRI mapping

Authors: *Q. ZHU[1], ***Q. ZHU**[1], W. VANDUFFEL[1,2,3]
[1]Lab. for Neuro- and Psychophysiology, KU Leuven, Leuven, Belgium; [2]Dept. of Radiology, Harvard Med. Sch., Boston, MA; [3]A.A. Martinos Ctr. for Biomed. Imaging, Massachusetts Gen. Hosp., Charlestown, MA

Abstract: The visuotopic organization of third tier visual areas in macaque monkeys is still heavily debated. One prominent model (Gattass et al., 2015; Kaas et al., 2015) proposed that V3 extends along most of the anterior V2 border, containing a split upper and lower visual field (VF) representation mirroring that of V2. Although this simple organization holds for V3v, V3d is more complex, with often a reported gap dividing it in two parts. Rostral to V3d, another third tier area (V3A) is described with both an upper (more caudally) and lower (rostral) VF representation, and a lower vertical meridian (VM) representing its anterior border with V4d. Another model (Angelucci and Rosa, 2015), however, considered V3d and the upper quadrant representation of V3A as part of area DM. The lower quadrant representation of V3A, on the other hand, is considered as a continuation of V3v. Together they form a complete representation of the VF similar to VLP of New World monkeys (marmosets). To evaluate these models, we acquired retinotopy maps from 3 awake rhesus monkeys using high resolution phase-encoded retinotopic fMRI mapping (0.6 mm isotropic voxels). To better detect fine grained VF transitions, we superimposed isopolar angle and isoeccentricity contour lines on field sign maps (Sereno et al., 1994). Furthermore, population receptive field (pRF) sizes (Dumoulin and Wandell, 2008) were calculated to quantitatively test the two models. Our results show that rostral to V2d there are two separated mirror field sign maps with a gap, exactly as described above for V3d. However, the more dorso-medial area shares a horizontal meridian with V2d, as V3d, and a lower VM with another area, while the more ventro-lateral area bends away from V2d, and contains a representation of both the upper and lower VF, very similar to DLP as described in New World owl monkeys (Sereno et al., 2015). This more ventral area, has the same non-mirrored field sign as V3v with which it forms a continuous band, but it is distinct from VLP because of the upper VF representation. The pRF size in this macaque counterpart of DLP is also significantly larger than that found for V3d and V3v -the two latter areas have the same pRF sizes. Therefore, DLP cannot be considered as another part of V3d, as proposed by the first model, nor the continuation of V3v, as proposed by the second model. Its pRF size, however, is similar to that of V4d and V4v, suggesting that DLP is a fourth tier area, consistent with the observation of Zeki and Baizer et al. that multiple VFs are represented in V4d (Zeki, 1971;

Baizer and Maguire, 1983). In summary, the retinotopic maps of dorsal visual cortex is more similar in Old and New World monkeys than generally appreciated.

Disclosures: Q. Zhu: None. **W. Vanduffel:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.05/II19

Topic: D.07. Vision

Support: Grant–in–Aid for JSPS Fellows (16J01822)

Title: Transient oscillatory feedback from ipsilateral IPS in response to a visual target

Authors: *K. YUASA[1,2,3], H. TAKEMURA[1,2,3], I. MOTOYOSHI[4], K. AMANO[1,2]
[1]CiNet, NICT, Osaka, Japan; [2]Grad. Sch. of Frontier Biosci., Osaka Univ., Osaka, Japan; [3]JSPS, Tokyo, Japan; [4]Dept. of Life Sci., The Univ. of Tokyo, Tokyo, Japan

Abstract: A number of experimental and theoretical studies suggest a fundamental role of intra-cortical feedback loops in visual processing (Hochstein & Ahissar, 2002; Lamme, 2001). Recent studies proposed that feedforward and feedback signals have been associated with different frequencies of neural oscillations based on macaque electrophysiology (van Kerkoerle et al., 2014) and human MEG (Michalareas et al., 2016). While these characteristics were mostly studied for late sustained responses, early transient responses are also crucial for visual processing. Here we investigated temporal property and stimulus dependency of feedforward and feedback signals in the early response to visual stimulus onset.
Twenty healthy participants viewed a monochromatic checker disc that was presented for 500 ms at one of four possible locations (upper-left, upper-right, lower-left, and lower-right) with respect to the central fixation dot. The stimulus contrast was either 100% or 30%. For each condition, MEG signals were recorded repeatedly over 216 trials with an inter-trial interval of 1000-1500 ms. The source activities were estimated by means of dSPM for twelve region-of-interests (ROIs; V1, V2v, V3v, hV4, VO/PHC, V2d, V3d, V3a/b, LO, TO, IPS0, and IPS/SPL) that were identified via surface-based fitting of retinotopy atlas (Wang et al., 2015). For every pair of ROIs, we applied a granger causality analysis in the time-frequency domain, and looked at feedforward and feedback interactions across cortical areas.
The analysis revealed strong feedforward signals from V1 to the other visual areas and feedback signals from the IPS/SPL to the other visual areas ($p<0.05$). Both were clearly observed in the beta band at 100-160 ms after the stimulus onset. The feedback signals were significantly larger in the ipsilateral hemisphere than in the contralateral hemisphere with respect to the stimulated visual field ($p<0.05$). While the evoked response increased with the stimulus contrast ($p<0.05$),

we did not find a significant contrast dependency in the amplitude of the feedforward and feedback signals ($p>0.05$).
Given the retinotopic organization of IPS, it has been suggested that some top-down modulation in the primary visual cortex is regulated by IPS (Silver & Kastner, 2009). Indeed, the parieto-occipital network in the contralateral hemifield has been considered critical for the control of spatial attention (Kastner et al., 1999; Saalmann et al., 2007). While dominant feedback from the ipsilateral IPS is apparently inconsistent with the previous studies, it may suppress signals in the ipsilateral visual field where the target is absent for efficient visual processing.

Disclosures: K. Yuasa: None. **H. Takemura:** None. **I. Motoyoshi:** None. **K. Amano:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.06/II20

Topic: D.07. Vision

Support: NSF EPSCoR RII Track-2 (T-2) award

Title: Dynamic lateral interactions in monkey area V4

Authors: *R. XIA[1], S. GUAN[2], D. L. SHEINBERG[3]
[2]Dept. of Neurosci., [3]Neurosci., [1]Brown Univ., Providence, RI

Abstract: The primate visual system has long been thought as a feedforward hierarchical machine with independent filtering nodes. Less is known about the dynamics and the role of lateral interactions within a visual region. To investigate the pattern of these recurrent interactions, we carried out multi-electrode recordings in monkeys' intermediate visual area V4 when monkeys were looking at visual stimuli or performing feature-based attention tasks. We found that the pairwise coherence between local field potentials was strongly modulated by task events. Despite the large variation in such modulations, our decoding results indicated that the synchrony pattern contained information about both visual and attentional features. Furthermore, Granger causality analysis showed asymmetric interactions between foveal and peripheral neurons. We found that the signal flow from peripheral channels to foveal channels were increased during full-screen visual presentation, whereas small central visual stimuli induced a larger increase of Granger influences from foveal channels to peripheral channels. In contrast, the local interactions within foveal and peripheral groups, but not the cross-group interactions, were enhanced during the feature attention-modulated delay period. Put together, our results suggested a task-related, receptive field-specific interaction pattern. In order to further probe the recurrent functional connectivity in V4 during visual attention tasks and also causally test its physiological and behavioral roles, our ongoing project involves multi-channel optogenetic

stimulation and simultaneous recordings. We hope that our data will provide more insights to the understanding of the dynamical, highly interactive visual network.

Disclosures: R. Xia: None. **S. Guan:** None. **D.L. Sheinberg:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.07/II21

Topic: D.07. Vision

Support: NIH RO1-EY014681

ONR N000014-14-0670

Title: Bidirectional visual processing: Distinct dynamics and interactions between V4 and inferior temporal cortex in challenging scenarios

Authors: *S. GUAN[1], R. XIA[2], D. SHEINBERG[2]
[1]Dept. of Neurosci., [2]Neurosci., Brown Univ., Providence, RI

Abstract: The feedforward hierarchical framework of the ventral stream greatly advances our understanding of visual objection recognition. However, there are as many feedback/horizontal projections as the feedforward ones, whose roles in visual recognition are largely unknown. This current study aims to investigate the visual processing beyond the feedforward sweep in a naturalistic setting. To engage the feedback/horizontal mechanisms, we probe the visual system using full screen natural images with two manipulations: 1) the images are either familiar (after months' exposure) or novel, and 2) the images are overlaid with different levels of occluding noise. Our behavioral data shows that recognition requires longer image presentation time when higher level of noise is added, and familiarity helps recognition in high noise conditions, which suggests the possible involvement of feedback processing. To find the neural correlate, we did simultaneous multi-channel electrophysiological recordings in areas V4 and inferior temporal cortex (IT) in a macaque monkey. We found that V4 and IT respond differentially to our image manipulations: 1) occluding noise only scales down the averaged population firing dynamics in V4, whereas it flattens the dynamics in IT, by reducing the early peak and increasing the late-phase response; 2) familiarity reduces the averaged population firing rate earlier and to a larger extend in IT than in V4; 3) population decoding results shows that occluding noise largely scales down the image information contained in V4, whereas it reduces the early phase of IT's information to a smaller extend but leave the late phase unaffected; 4) functional connectivity analysis using local field potential Granger causality suggests that feedforward information flow from V4 to IT is largely reduced by noise, whereas the feedback information flow from IT to V4

is not affected by noise, and emerged later than the feedforward flow. Taken together, we our results provide evidence for the bidirectional visual recognition processing in a challenging naturalistic scenario.

Disclosures: **S. Guan:** None. **R. Xia:** None. **D. Sheinberg:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.08/II22

Topic: D.07. Vision

Support: BBSRC

The Royal Society

Wellcome Trust

Title: Distinct intrinsic connectivity patterns for dorsal and ventral regions of lateral intraparietal area (LIP) of macaque monkeys

Authors: *M. RUESSELER[1], B. AHMED[1], J. BENNETT[1,2], J. E. T. SMITH[1], A. J. PARKER[1], K. KRUG[1]
[1]Dept. of Physiology, Anat. and Genet., Univ. of Oxford, Oxford, United Kingdom; [2]Dept. of Informatics, Univ. of Sussex, Falmer, Brighton, United Kingdom

Abstract: Neurons in parietal area LIP respond to visual stimuli and serve to integrate visuo-motor information to an attended saccade location. Dorsal (LIPd) and ventral (LIPv) regions can be distinguished based on myelination, cytoarchitecture and inter-areal connectivity. Here, we examine the intrinsic pattern of local connections of LIPd/LIPv by analysing the distribution of labelled cells following focal tracer injections.
Using structural MRIs under Brainsight (Rogue Research Inc) control, we injected (~80 nL) retrograde tracer into LIP of four male Rhesus macaques (*Macaca mulatta*). Three animals received LIPd injections (Cholera-Toxin b (CTb) N=2; Fluorogold (FG) N=1); one received a CTb injection into LIPv. From a 1:5 series of 50 µm parasagittal sections, we marked labelled cell bodies with Neurolucida (Microbrightfield Ltd). Cell density was quantified by smoothing cell locations with a Gaussian spread (sd = 100µm); distinct clusters of high-density label were identified (contours at >90% of max density). For two animals with CTb injections in LIPd, we generated a 2D spatial map of cell density distribution. A polynomial described the distribution of labelled cells separately for superficial and deep layers in each section (fitted R^2 >=0.9): locations of cells were projected onto this line to measure the variation in density of labelled cells along the dorso-ventral extent of LIP. Consecutive parasagittal sections were aligned based

on nearest neighbour density distributions of labelled cells.
Injection in LIPd gave a widespread pattern of labelled cells within LIPd, for all layers. Along the dorso-ventral extent, the density distribution formed a longitudinal patch of labelled cells centred on the location of the injection site. Along the medio-lateral axis, a small number of large but distinct patches of labelled cells were found about 2 to 3 mm away from the injection site. Within single sections, cell density analysis indicated a more fine-grained organization, with distinct smaller clusters of labelled cells with the majority of intercluster distances between 0.5 and 1mm. In stark contrast, in LIPv only one cluster of labelled cells was found after tracer injections in LIPd. Conversely, data from the single animal with an injection in LIPv revealed a more widespread pattern of retrogradely labelled cells spanning both LIPv and LIPd.
The widespread connectivity pattern found within LIPd suggests that a number of distant visual field locations might contribute to receptive/response field properties of cells. The restricted point-to-point projection from LIPv to LIPd might relate to the structure of the visuotopic map previously shown to exist in LIPv.

Disclosures: **M. Ruesseler:** None. **B. Ahmed:** None. **J. Bennett:** None. **J.E.T. Smith:** None. **A.J. Parker:** None. **K. Krug:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.09/II23

Topic: D.07. Vision

Support: NIMH IRP (MH002909-08)

Title: Foveal, elliptical and horizontal - Population receptive fields in the visual word form area are tuned optimally for word reading

Authors: *E. H. SILSON[1], R. C. REYNOLDS[2], D. JANINI[1], C. I. BAKER[3], D. J. KRAVITZ[4]
[1]Lab-Brain & Cog, [2]Scientific and Statistical Computing Core, Natl. Inst. of Mental Hlth., Bethesda, MD; [3]Lab. Brain and Cognition, NIH, Bethesda, MD; [4]Dept. of Psychology, The George Washington Univ., Washington, DC

Abstract: Our ability to identify words is optimal at fixation and deteriorates rapidly with increasing eccentricity. In cortex, word reading is thought to be subserved by a specialized region, the visual word form area (VWFA), in left ventral occiptotemporal cortex. Despite early assertions of position invariance, recent work demonstrates position sensitivity within VWFA (Rauschecker et al., 2012; Le et al., 2017).
Here, using a population receptive field (pRF) model that allows for estimates of elliptical and orientated pRFs, we investigated the pRF properties of VWFA. Specifically, we tested the

prediction that pRFs in VWFA would be 1) foveally centered, 2) highly elliptical and 3) horizontal - the ideal parameters for recognizing word forms in languages that read left-right. Importantly, we contrast the pattern of VWFA pRFs with those exhibited by two control regions in which we do not predict this specific pRF combination: the parahippocampal place area (PPA), and early visual cortex (EVC) V1-V3.
Thirteen participants completed eight pRF runs. In each run, a bar aperture revealing scene fragments traversed the visual field, whilst participants fixated centrally. Our VWFA definition was taken from the group average in Stevens et al (2017), whereas PPA and EVC were identified functionally in each individual.
We computed distributions of eccentricity, aspect ratio, and orientation theta in each participant and region, respectively. First, consistent with our predictions, the distribution of eccentricity within VWFA peaked within the first two degrees of visual angle, which was significantly more foveal than that of either PPA or V1. Second, VWFA showed significantly more elliptical pRFs than either PPA and V1; VWFA showed a distribution skewed significantly towards greater ellipticities. Third, unique to VWFA, pRF orientations were tightly distributed around horizontal. To confirm this specific pRF combination in VWFA, we further identified and measured VWFA pRFs in an additional six participants at high-resolution ($1.2mm^3$). The patterns of results were indistinguishable from our group-based approach. As with the group-based ROIs, pRFs in individually defined VWFAs were foveal, elliptical, and horizontal. Our data suggests two possibilities. First, pRFs in VWFA are randomly tuned to these characteristics and that guides the formation of the VWFA. Second, and more likely, is that learning to read shapes pRFs in VWFA, making them ideal for word reading. What form the pRFs in this region take in children prior to reading, in dyslexic individuals, or readers of languages not written from left to right are critical questions for the future.

Disclosures: **E.H. Silson:** None. **R.C. Reynolds:** None. **D. Janini:** None. **C.I. Baker:** None. **D.J. Kravitz:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.10/II24

Topic: D.07. Vision

Support: PFV/10/008

Fonds voor Wetenschappelijk Onderzoek

Title: Multi-electrode recordings in human lateral occipital cortex

Authors: *E. PREMEREUR[1], T. DECRAMER[2], W. VAN PAESSCHEN[1], P. JANSSEN[1], J. VAN LOON[3], T. THEYS[4]
[2]Lab. voor Neuro- en Psychofysiologie, [1]KU Leuven, Leuven, Belgium; [3]UZ Leuven, Leuven, Belgium; [4]Lab. Exp. Neurochirurgie En Neuroanatomie, Leuven, Belgium

Abstract: The lateral occipital complex (LOC) is a set of areas in human occipito-temporal cortex responding more to objects as opposed to non-object control stimuli, but the correspondence between LOC and the macaque inferotemporal areas is poorly understood. We had the unique opportunity to implant a 96-channel Utah array in human LOC, in a patient requiring surgery for epilepsy, at MNI coordinates -50 (mediolateral), -77 (anteroposterior), 6 (dorsoventral). An LOC localizing fMRI experiment performed after array extraction showed stronger activations during presentation of non-scrambled versus scrambled objects at the implantation site. Multi-unit (MUA) and local field potential (LFP) activity was recorded on four consecutive days, and the LOC localizer task was presented to the patient on three out of four days (day 1, 2 and 4). During the LOC localizer task, we presented images of stimuli, stimulus-outlines and scrambled controls for both, as in previous fMRI experiments, while monitoring the position of both eyes using an infrared tracker (Iscan). We found significant stimulus-induced gamma (25-150 Hz) responses (shapes compared to baseline) on all channels (n = 95) in all three sessions (permutation test, $p < 0.01$). Moreover, 92 (day 1), 55 (day 2) and 79/96 (day 4) channels showed significant differences in gamma response between scrambled and non-scrambled objects. Receptive field testing showed significant gamma responses during ipsi- and contralateral stimulus presentation on virtually all channels (permutation test, $p < 0.01$). We obtained qualitatively similar results when restricting the analysis to the high-gamma band (80-120 Hz). Furthermore, we measured significant MUA spiking responses during visual stimulation compared to baseline on 19 channels, and significant differences in MUA response between non-scrambled and scrambled stimuli on 9 channels (day 1). MUA latencies were around 175 msec, with a peak response at 200 msec. As a final test, we recorded neural activity during foveal presentation of disparity-defined convex or concave stimuli on a stereoscope during binocular eye movement control. We found an increase in gamma power on 92 channels (permutation test, $p < 0.01$) during stimulus presentation compared to baseline. Importantly, 11/95 channels on day 1 and 7/95 channels on day 2 showed a significant preference in gamma response to convex versus concave stimuli, which was preserved across positions in depth (ANOVA, significant main effect of stereo; no significant interactions), indicating higher-order disparity selectivity. Taken together, these results suggest that human LO may correspond more to macaque area TE than to area TEO.

Disclosures: E. Premereur: None. **T. Decramer:** None. **W. Van Paesschen:** None. **P. Janssen:** None. **J. Van Loon:** None. **T. Theys:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.11/II25

Topic: D.07. Vision

Support: NIH 1R01EY02391501A1

NIH RO1EY02988-A1

Title: Cortical thinning of functional regions in the ventral temporal cortex is associated with increased myelination

Authors: *V. S. NATU[1], J. GOMEZ[2], M. BARNETT[1], B. JESKA[1], Z. ZHEN[1], S. COX[1], K. GRILL-SPECTOR[3,4]
[1]Dept. of Psychology, Stanford University,, Stanford, CA; [2]Neurosciences Program, Stanford Univ. Sch. of Medicine, Stanford, Stanford, CA; [3]Stanford Univ., Stanford, CA; [4]Stanford Neurosciences Institute, Stanford Univ., Stanford, CA

Abstract: The human brain undergoes substantial anatomical changes from childhood to adulthood including decreases in gray matter volume, cortical thinning (Sowell et al., 2004), and increases in white matter volume (Giedd et al., 1999). However, the mechanisms underlying these developments, especially the thinning of cortex, are unknown. We tested two main hypotheses of cortical thinning: pruning and growth. Pruning suggests that cortical thickness (CT) decreases due to reduction in tissue volume from childhood to adulthood. Thus, pruning predicts higher T_1 relaxation rate, measured with quantitative MRI (qMRI), and higher mean diffusivity (MD), measured with diffusion MRI (dMRI), in adults than children. In contrast, growth associated with increased myelination, predicts lower T_1 and lower MD in adults than children. We tested these hypotheses using fMRI, qMRI, and dMRI in 27 children (ages 5-9, *N*=12; ages 10-12, *N*=15) and 30 adults (ages 22-28). We identified face-, body-, character- and place-selective regions in ventral temporal cortex (VTC) of each participant and measured in each region CT, T_1, and MD, as well as T_1 and MD in the neighboring white matter. We observed four main findings: First, CT significantly decreased from childhood to adulthood in all category-selective regions (Fs>27.67, ps<0.001). The cortex thinned on average by 0.8±0.18 mm, with the least thinning in the right place-selective region. Second, in regions that we observed significant reduction in CT, we also found significant reduction in T_1 from childhood to adulthood ($F_{2,492}$=12.75, p<0.001). Third, both T_1 and MD of white matter neighboring the face-, body- and character-selective (but not place-selective) regions showed significant age-related reductions (Fs>35.22, ps<0.001). Fourth, analysis of T_1 and MD by cortical layers showed that greatest developmental changes occur close to the gray-white matter boundary and the developmental shifts in T_1 curves by layer is similar to the developmental changes observed in

cortical thickness. Together, our data (i) reveal differential development of tissue properties across human VTC, and (ii) provide strong evidence that cortical thinning is associated with increased myelination in deep cortical layers and adjacent white matter, rather than pruning. As developmental changes in CT are prevalent throughout the brain, our results have important implications for interpreting the mechanisms of CT during typical and atypical development.

Disclosures: V.S. Natu: None. **J. Gomez:** None. **M. Barnett:** None. **B. Jeska:** None. **Z. Zhen:** None. **S. Cox:** None. **K. Grill-Spector:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.12/II26

Topic: D.07. Vision

Support: Fonds voor Wetenschappelijk Onderzoek and PFV/10/008.

Title: Functional magnetic resonance imaging (fMRI) - Guided single unit recordings reveal weak higher-order disparity selectivity in macaque area TEO

Authors: *A. ALIZADEH, R. VOGELS, P. JANSSEN
Katholieke Univ. Leuven, Leuven, Belgium

Abstract: Binocular disparity provides a strong, unambiguous cue for depth perception in primates. Several cortical areas in both the dorsal and the ventral visual pathways are involved in the processing of depth from binocular disparity. Previous functional magnetic resonance imaging (fMRI) studies in monkeys (Van Dromme et .al, PLoS Biol. 2016 Apr 15;14(4):e1002445. doi: 10.1371) revealed stronger activations in the posterior part of the inferotemporal cortex (area TEO) in response to higher-order (curved and slanted) disparity stimuli compared to zero-order (flat) stimuli. The current study was carried out to investigate the higher-order disparity selectivity in area TEO at the single-cell level. In two monkeys, we targeted the fMRI activation elicited by curved surfaces in area TEO during a passive fixation task. The search test consisted of 32 second-order stimuli in addition to 8 first-order stimuli (2 vertical and 2 horizontal linear disparity gradients, square shape, 4 of which were 8.4° and the other 4 were 18.75° in diameter). We tested for disparity selectivity (including monocular controls) and selective cells were additionally tested with preferred and non-preferred stimuli at 5 different positions in depth (mean disparity from -0.5 to 0.5 deg). In total, we recorded from 123 TEO neurons showing significant visual responses to the stimuli of the search test presented at the fixation point. A small fraction of all visually responsive neurons (20 neurons, 16%) exhibited significant selectivity for disparity-defined curved and slanted stimuli, which could not be explained by the monocular responses. Moreover, the weak disparity selectivity of TEO

neurons emerged very late after stimulus onset (150 ms). Only four neurons (3% of all visually responsive neurons) preserved their selectivity across positions in depth when tested with second-order (curved) stimuli. .Disparity-selective TEO neurons usually had small receptive fields (average RF size = 32 deg^2) centered on the fovea. Our results suggest that the fMRI activations observed in TEO overestimates the neuronal selectivity for disparity-defined curved surfaces. The weak disparity selectivity we measured in TEO may represent feedback signals from higher-order areas in the ventral visual stream.

Disclosures: **A. Alizadeh:** None. **R. Vogels:** None. **P. Janssen:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.13/II27

Topic: D.07. Vision

Support: NIMH intramural research funding

Title: Categorization in monkey inferior temporal cortex determined by image features, not acquired knowledge

Authors: *X. YUE[1], M. YETTER[2], L. G. UNGERLEIDER[3]
[2]Natl. Inst. of Mental Hlth., [1]NIH, Bethesda, MD; [3]Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: Our visual system represents visual objects according to categories. One of most basic categories is the animate vs. inanimate division. Based on results from recent studies (Livingston et al., 2015; Perrinet & Bednar, 2015) and our earlier study (Yue et al., 2014), we hypothesized that animate vs. inanimate categorization is: 1) encoded by multivoxel activity patterns; and 2) influenced to a large degree by the unique image-based features that distinguish animate from inanimate stimuli. Using a slow event-related design, we acquired fMRI scans in three fixating rhesus macaques in response to a large set of visual stimuli, including 47 sub-categories with 20 images per sub-category. We employed curved and rectilinear Gabor filters to quantify curved and rectilinear image-based features. As hypothesized, we found that multivoxel activity patterns measured with support vector machine classification encoded animate vs. inanimate categories in the monkey inferior temporal cortex; multiple dimensional scaling failed to categorize individual exemplars in the animate vs. inanimate division. This result suggests that animate vs. inanimate categorization is represented in the brain in a high dimensional space, and is not a 2-dimensional representation. Moreover, curved and rectilinear features explained a significant amount of variance in the fMRI activity patterns that encoded animate vs. inanimate categories. Our results thus support our hypothesis that animate vs. inanimate categorization in the inferior temporal cortex is influenced to a large extent by the unique image-based features (such as curved and

rectilinear features) that distinguish animate vs. inanimate stimuli. The results argue against the notion that categorization stems from acquired semantic knowledge of the characteristics that distinguish object categories, and instead suggest that the unique image-based features that distinguish animate vs. inanimate stimuli give rise to the formation of categorization in the macaque inferior temporal cortex.

Disclosures: X. Yue: None. **M. Yetter:** None. **L.G. Ungerleider:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.14/JJ1

Topic: D.07. Vision

Support: Conte (P50 MH942581A)

HHMI

Title: Principles of object representation in two object networks in IT cortexprinciples of object representation in two object networks in IT cortex

Authors: *P. BAO[1], L. SHE[2], M. MCGILL[3], D. Y. TSAO[2]
[1]Biol., [3]Computation and Neural Systems, [2]Caltech, Pasadena, CA

Abstract: Understanding the neural mechanism of object recognition is one of central challenges in visual neuroscience. Last year at SFN, we reported discovery of a new “object network” in macaque IT cortex consisting of three connected, discrete patches of cells with highly consistent response selectivity to a set of objects (Bao & Tsao, 2016). Here, we further probed the coding scheme used by neurons in this object network. For comparison, we also probed neurons in a body network. We performed extracellular recordings in both the object and body networks while monkeys kept fixation for juice reward and viewed a stimulus set consisting of realistic objects and their silhouettes. A large proportion of neurons in both the object and body networks showed indistinguishable responses to the real objects and their silhouettes. This suggests that we can reasonably reduce the real object image space to a much simpler silhouette space. To explore how neurons code the silhouette space, we recorded responses of cells in the middle body patch and anterior object patch to 2000 silhouette images randomly sampled from a 30 dimensional (30-d) space, extracted from a large set of object silhouette images by principal components analysis. We then fed the same image sets (real objects, silhouettes of real objects, silhouette images sampled from 30-d space) into a trained deep network, Alexnet, and fitted actual neuronal responses to the 2000 random silhouettes to a linear combination of artificial neurons’ responses from AlexNet. We found this linear model could effectively predict responses of

neurons to the real object silhouette images as well as to the original images, with correlation between predicted and actual neuronal responses as high as the noise ceiling. Overall, these results show that we can model the object code used in the macaque object and body networks with high precision, and that at least within these two networks, this code largely based on silhouettes of objects.

Disclosures: **P. Bao:** None. **L. She:** None. **M. McGill:** None. **D.Y. Tsao:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.15/JJ2

Topic: D.07. Vision

Support: HHMI

Title: The role of disparity columns in segmentation

Authors: ***J. K. HESSE**[1], D. Y. TSAO[2]
[1]Biol., [2]Caltech, Pasadena, CA

Abstract: Segmentation is a process crucial for interacting with objects in the world, as one needs to know what regions of visual space constitute the objects that we interact with and where the borders are that belong to them. We set out to find out whether there exist specialized regions that are responsible for segmentation. Disparity columns in V3 found by Adams and Zeki (J Neurophys, 2001) contain alternating clusters of cells tuned to far and near disparity, respectively. Since disparity information is an important cue for interpreting 3D objects and surfaces, we hypothesized that disparity columns might play a role in segmentation, too. We performed tangential recordings along the fundus of the lunate sulcus and found that, besides clusters of cells tuned to near and far, disparity columns also contain clusters of cells tuned to the border-ownership of a disparity-defined square. We found that these cells encoding disparity-defined borders also carried significantly more segmentation-related information for squares defined by other modalities, such as luminance, texture, motion and illusory contours.
Our results suggest that disparity-columns in V3 may serve a broader role rather than just computing disparity and may be responsible for segmenting the visual scene into surfaces corresponding to objects.

Disclosures: **J.K. Hesse:** None. **D.Y. Tsao:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.16/JJ3

Topic: D.07. Vision

Support: Simons foundation grant ID# 325023

Swartz grant # 2013-36

HHMI

Title: Coding of 3d-surfaces by neurons in the primate caudal intraparietal sulcus (CIPS)

Authors: *L. CHANG[1], D. Y. TSAO[2]
[1]Div. of Biol., CALIFORNIA INSTITUTE OF TECHNOLOGY, Pasadena, CA; [2]Caltech, Pasadena, CA

Abstract: The central function of the visual system is to infer events happening in the 3-d world. One key step of this inference is to extract depth/disparity information from 2-d inputs. Previous fMRI studies identified an area in the parietal lobe, CIPS (caudal intraparietal sulcus), which responds selectively to images defined by variations in disparity (Tsao et al., 2003). Electrophysiology revealed that neurons in this area are tuned to 3-d features such as tilt and slant of surfaces (Tsutsui et al., 2001, Rosenberg et al., 2013). However, the full mechanism by which CIPS neurons encode 3-d structure remains unclear. We targeted CIPS for electrophysiology recording, guided by fMRI scans using stereo images. Tilting surfaces similar to a previous study (Rosenberg et al., 2013) were first presented to determine the preferred orientation of the recorded neuron. Surfaces with depth varying as a sinusoidal function along this orientation or along an axis orthogonal to this orientation were synthesized and presented. We found CIPS neurons displayed diverse tuning to this stimulus set. To further explore the effective features coded by each neuron, the sinusoidal stimuli at the preferred spatial frequency and orientation were sectioned into smaller segments. As a result, the new stimulus set contained different kinds of shapes (slanting surfaces, convex or concave surfaces) at varying locations. We found two types of behavior in the recorded neurons. Type 1 neurons preferred the same feature at different locations, while type 2 neurons preferred different features at different locations. Further analyses revealed that responses of type 2 neurons could be explained by a receptive field structure analogous to simple cells in V1: instead of coding different polarity in light intensity as in V1 simple cells, the type 2 neurons code different polarity in depth/disparity at different 2-d spatial locations.

Disclosures: L. Chang: None. **D.Y. Tsao:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.17/JJ4

Topic: D.07. Vision

Title: Receptive fields of adjacent neurons in primate prefrontal and parietal cortices

Authors: *P. VISWANATHAN[1,2], A. NIEDER[1]
[1]Inst. of Neurobiology, Univ. of Tuebingen, Tübingen, Germany; [2]IMPRS for Cognitive and Systems Neurosci., Tuebingen, Germany

Abstract: The basic organization principles of the primary visual cortex (V1) are commonly assumed to also hold in the association cortex such that neurons within a cortical column share functional connectivity patterns and represent the same region of the visual field. Adjacent neurons within these cortical columns show nearly identical receptive field (RF) locations and stimulus tuning properties. We examined this principle in the prefrontal cortex (PFC) and ventral intraparietal area (VIP) of two rhesus monkeys by mapping the visual receptive fields of neurons recorded at the same electrode. By first creating high-resolution RF maps for the neurons that selectively responded to the position of a moving bar stimulus and comparing those of neuron pairs, we found that the spatial characteristics of visual receptive fields between adjacent neurons differed considerably, with increasing heterogeneity from VIP to PFC. To further examine how the fine-scale specificity of connections between adjacent neurons might explain the relationship between their receptive fields, we performed cross-correlation analysis of their spike trains. In addition to receptive field incongruences, putative inhibitory interneurons and pyramidal cells were differentially connected in the two areas, with inhibitory interactions much more prevalent between PFC neuron pairs than those in VIP. These findings suggest that local receptive field topography vanishes with hierarchical distance from visual cortical input and argue for increasingly modified functional microcircuits in non-canonical association cortices that contrast V1.

Disclosures: P. Viswanathan: None. **A. Nieder:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.18/JJ5

Topic: D.07. Vision

Support: NIH R01 EY023384

Title: Voxel-to-voxel encoding models of connectivity between visual areas

Authors: *M. MELL, T. P. NASELARIS
Med. Univ. of South Carolina, Charleston, SC

Abstract: In the human brain, vision is accomplished collectively by a multitude of interconnected but distinct cortical areas. We are still in the early stages of our understanding of the way that connections between these areas enable visual perception. In particular, we know very little about the functional differences between feedforward, feedback, and within-area connections. We developed a voxel-to-voxel encoding model approach to quantify the spatial patterns and predictive accuracy of the three types of connections across visual brain areas. In an fMRI experiment we measured BOLD responses to natural images in seven visual areas treated in our study as an ordered, hierarchical sequence: V1, V2, V3, V4, V3A, V3B, and the lateral occipital complex (LO). We then constructed encoding models that predicted the activity variation in one voxel as a function of the activity variation in other voxels. For each voxel, we constructed seven distinct encoding models. Each model predicted the target voxel's activity as a function of activity in one of the seven visual areas (including the visual area occupied by the target voxel). Models that connect lower to higher visual areas (e.g., V1 to LO) were considered feedforward. Models connecting higher to lower areas (e.g., LO to V1) were considered feedback. Models connecting voxels in the same areas were considered internal. For each type of encoding model we measured cross-validated prediction accuracy. Several interesting trends emerged from this analysis. First, we found that for purely linear feedforward encoding models prediction accuracy decayed rapidly across the visual hierarchy from V1 to LO. Prediction accuracy for linear internal encoding models also decayed rapidly. However, for linear feedback models prediction accuracy showed little or no decay across cortical areas, and was roughly matched to the accuracy of the internal model. When nonlinear encoding models (implemented as a shallow neural network) were applied, the asymmetry in the pattern of prediction accuracy between feedforward and feedback models was preserved; however, the prediction accuracy for the internal models was markedly improved, and remained stable across visual areas. A preliminary interpretation of these results is that connectivity in the feedforward direction is nonlinear at each stage (as in a deep neural network), compounding the failure of purely linear, or nonlinear but shallow models, across the cortical hierarchy. Connections in the feedback direction appear not to accumulate nonlinearities at each stage, explaining why the performance of purely linear models was independent of distance along the cortical hierarchy.

Disclosures: M. Mell: None. **T.P. Naselaris:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.19/JJ6

Topic: D.07. Vision

Support: European Union's Horizon 2020 research and innovation programme under the Marie Sklodowska-Curie Grant agreement No 641805

National MS society research grant 5128-A-1

Title: The effect of ocular dominance and binocularity on population receptive fields' properties

Authors: *P. B. DE BEST, N. RAZ, N. LEVIN
fMRI unit, Neurol. Dept., Hadassah Hebrew Univ. Med. Ctr., Jerusalem, Israel

Abstract: *Introduction:* In the seminal Hubel & Wiesel (1959, 1962) work in cats, ocular dominance and binocularity effects on visual perception were assessed. **Ocular dominance**, the tendency to prefer visual input from one eye to the other was found to be the main factor contributing to receptive field differences between the eyes. **Binocular** stimulation resulted in a stronger response than monocular stimulation, a phenomenon known as binocular summation. Herein, we use the population receptive field (pRF) method to study the neural representation of monocular and binocular vision to better understand the origin of these phenomena.
Methods: Thirteen subjects were scanned in a 3T Siemens Skyra magnetic resonance imaging (MRI) scanner while viewing drifting bar stimuli and performing a fixation task. Both eyes (binocular condition), the dominant eye and the non-dominant eye (two monocular conditions) were stimulated in separate sessions. The pRF size and amplitude were compared between the three conditions along the eccentricity axis. Binocular summation ratios were calculated by dividing binocular by mean monocular amplitude and pRF size.
Results: To evaluate viewing condition and experimental order effects (e.g. running the binocular condition first or last), analysis of variance (ANOVA) was performed. A main effect for the tested eyes condition (dominant, non-dominant or binocular) was found in V2 at 5-6^{o} eccentricity. Post-hoc tests indicated that pRF size was significantly larger for binocular than for non-dominant eye conditions. In addition, significant interactions between condition and order were found in lower eccentricities, suggesting that binocularity may be affected by prior exposure to monocular stimulation.
It was also found that binocular amplitudes were significantly higher than monocular amplitudes, indicating a binocular summation effect. This effect occurred consistently, and varied between 1.1 and 1.3 among visual areas and across eccentricities. In contrast, pRF size summation only occurred at high eccentricities in V2, and probably resulted from the significant effect on pRF size described above.

Conclusions: Our results suggest that binocular pRF sizes in early visual areas are derived from the dominant, and not the non-dominant eye but no significant difference between the dominant and non-dominant eye in regard to the pRF size was found. Interactions with experimental order indicate that binocular pRF size may be influenced by prior exposure to monocular stimulation. The found summation ratios indicate that binocular amplitude summation occurs, despite a lack of pRF size differences between binocular and monocular conditions.

Disclosures: P.B. De Best: None. **N. Raz:** None. **N. Levin:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.20/JJ7

Topic: D.07. Vision

Title: Visual field map clusters in higher-order visual processing: Organization of visual field maps within the human lateral occipital cortex

Authors: *A. A. BREWER[1], B. BARTON[2]
[1]Cognitive Sci., UC Irvine, Irvine, CA; [2]Univ. of California Irvine, Irvine, CA

Abstract: Over the last ten years, the lateral part of the occipital pole has often seemed to be the Wild West of visual field mapping, with a variety of claims regarding retinotopic organization staking out this region. Studies have variously proposed that the lateral occipital cortex between V3d dorsally and hV4 ventrally and posterior to hMT+ has no retinotopic organization (Ishai et al., 2000; Kanwisher et al., 1997; Halgren et al., 1999), bears only a gradient of 'eccentricity bias' (Malach et al., 2002), or, ventral to LO-2, shows sensitivity to retinal position suggestive of underlying retinotopic representations (Sayres & Grill-Spector, 2008). More recent studies have begun to demonstrate VFMs in some sections of lateral occipital cortex, with somewhat contradictory reports of 2 dorsal VFMs (LO-1, LO-2; Larsson & Heeger, 2006) with non-orthogonal (15.3° offset) eccentricity and polar angle gradients and a shared polar angle border with hMT+/TO VFMs (Amano et al., 2009), or 2 dorsal VFMs now with orthogonal gradients (LO-1, LO-2) and a shared eccentricity border with hMT+/TO VFMs and 2 ventral VFMs organized into a cloverleaf cluster (phPIT; Kolster et al., 2010). Our results resolve these conflicting reports and extend the orthogonal VFM measurements by demonstrating that the lateral occipital cortex is home to 6 VFMs bilaterally, LO-1, LO-2, LO-3, LO-4, LO-5, and LO-6, which together form the lateral part of the occipital pole visual field map cluster. The peripheral eccentricity representations of the LO maps reverses anteriorly into the peripheral eccentricity representations of the hMT+/TO visual field map cluster. Reliability measurements of these VFMs reveal that these cloverleaf clusters are remarkably consistent and functionally differentiable. Our findings add to the growing number of measurements of widespread sensory

cortical field maps (CFMs) organized into cloverleaf clusters (e.g., Barton et al., 2012), indicating that CFMs and cloverleaf clusters may both be fundamental organizing principles in cortical sensory processing.

Disclosures: A.A. Brewer: None. **B. Barton:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.21/JJ8

Topic: D.07. Vision

Support: NSF Research Grant 1329255

Title: Visual field map clusters in high-order visual processing: Organization of V3A/V3B and a new cloverleaf cluster in the posterior superior temporal sulcus

Authors: *B. BARTON, A. A. BREWER
Univ. of California Irvine, Irvine, CA

Abstract: The cortical hierarchy of the human visual system has been shown to be organized around retinal spatial coordinates throughout much of low- and mid-level visual processing. These regions contain visual field maps (VFMs) that each follow the organization of the retina, with neighboring aspects of the visual field processed in neighboring cortical locations. On a larger, macrostructural scale, groups of such sensory cortical field maps (CFMs) in both the visual (Brewer et al., 2005; Wandell et al., 2005; Kolster et al., 2010) and auditory systems (Barton et al., 2012) are organized into roughly circular cloverleaf clusters. CFMs within clusters tend to share properties such as receptive field distribution, cortical magnification, and processing specialization. Here we use fMRI and population receptive field (pRF) modeling to investigate the extent of VFM and cluster organization with an examination of higher-level visual processing in temporal cortex and compare these measurements to mid-level visual processing in dorsal occipital cortex. In human temporal cortex, the posterior superior temporal sulcus (pSTS) has been implicated in various neuroimaging studies as subserving higher-order vision, including face processing (e.g., Hoffman and Haxby, 2000), biological motion perception (e.g., Grossman and Blake, 2002), and multimodal audiovisual integration (e.g., Beauchamp et al., 2004). In human dorsal occipital cortex, the transverse occipital sulcus (TOS) contains the V3A/B cluster, which comprises 2 VFMs subserving mid-level motion perception and visuospatial attention (Press et al., 2002; Smith et al. 1998). For the first time, we present the organization of VFMs in pSTS in a cloverleaf cluster. This pSTS cluster contains four VFMs bilaterally: pSTS-1:4. We characterize these pSTS VFMs as relatively small at ~125 mm^2 with relatively large pRF sizes of ~2-8° of visual angle across the central 10° of the visual field. V3A

and V3B are ~230 mm^2 in surface area, with pRF sizes here similarly ~1-8° of visual angle across the same region. In addition, cortical magnification measurements show that a larger extent of the pSTS VFM surface areas are devoted to the peripheral visual field than those in the V3A/B cluster. Reliability measurements of VFMs in pSTS and V3A/B reveal that these cloverleaf clusters are remarkably consistent and functionally differentiable. Our findings add to the growing number of measurements of widespread sensory CFMs organized into cloverleaf clusters, indicating that CFMs and cloverleaf clusters may both be fundamental organizing principles in cortical sensory processing.

Disclosures: B. Barton: None. **A.A. Brewer:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.22/JJ9

Topic: D.07. Vision

Support: (NWO) Vidi grant 452-08-008 to SD

Title: The fMRI contrast response function to natural images is enhanced according to local subjective importance

Authors: *W. ZUIDERBAAN[1], S. O. DUMOULIN[2]
[1]Utrecht Univ., Utrecht, Netherlands; [2]Spinoza Ctr. For Neuroimaging, Amsterdam Zuidoost, Netherlands

Abstract: INTRODUCTION
Our visual percept is not solely based on the sensory information that we receive from the light falling on our retina, but is also influenced by our knowledge of the world. There is no consensus about how this interaction influences the neural signal in early visual areas. Current theories implement this knowledge of the world as a perceptual hypothesis that is compared with the sensory input. According to different inference theories, the perceptual hypothesis can either be suppressing or boosting the sensory information represented by early visual areas. In our study we investigated the effect that the knowledge-based perceptual hypothesis has on the neural signal in early visual cortex and how this interacts with the responses to the sensory-driven image feature contrast.
METHODS
We measured the responses using 7T fMRI to different sets of stimuli: standard mapping stimuli, 45 natural images and a full-field stimulus (100% contrast).
The mapping stimuli were used to estimate the region of visual space a voxel responds to: the population receptive field (pRF, Dumoulin et al, 2008). We used a 2D Gaussian in the visual

field to represent the pRF.
We used the pRFs to quantify both the amount of RMS-contrast (sensory-driven property) and the amount of subjective importance (knowledge-based property) in the pRF.
To show how the neural responses are modulated by the RMS-contrast inside the pRF, we derived the contrast response function (CRF) by plotting the fMRI response amplitude against the RMS-contrast of all pRFs within a given visual field map.
The natural images were taken from the 'Berkeley Segmentation Dataset and Benchmark' database (Martin et al, 2001). In this dataset, for every image observers labeled the subjective important parts of the image. We use these manual labels to define and quantify our measure for the perceptual hypothesis in the pRF. Last, we show how the CRF is modulated by the amount of subjective importance inside the pRF.
RESULTS
Based on the inherent variations of contrast in the natural images we show that we can derive the contrast response function (CRF). Furthermore, we show how the CRF is modulated by the perceptual hypothesis as we defined it by subjective importance. The CRF was boosted in visual areas V1-V2-V3 when the responses were elicited by parts of the image of high subjective importance.
DISCUSSION
Thus, the sensory-driven image representation of a scene in early visual areas is boosted by the knowledge-based perceptual hypothesis. This result argues against most conventional implementations of predictive coding.

Disclosures: W. Zuiderbaan: None. **S.O. Dumoulin:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.23/JJ10

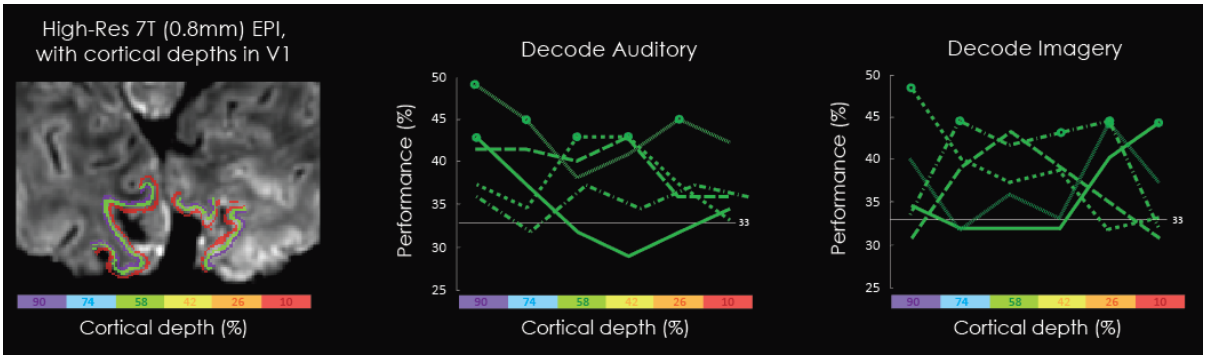
Topic: D.07. Vision

Support: ERC 2012 EtG 311751 BrainReadFBPredCode - 167640-01

Title: High-resolution 7T fMRI reveals auditory and imagery information in non-stimulated visual cortex

Authors: *M. BENNETT[1], L. S. PETRO[1], A. A. MORGAN[1], F. DE MARTINO[3], L. MUCKLI[2]
[1]Ctr. for Cognitive Neuroimaging, [2]Univ. of Glasgow, Glasgow, United Kingdom; [3]Fac. of Psychology, Maastricht Univ., Maastricht, Netherlands

Abstract: Introduction Feedback outnumber feedforward connections in human early visual cortex. Vetter, Smith and Muckli (2014) showed that complex auditory scenes played to blindfolded subjects can be classified from V1, V2 and V3 using multivariate techniques. This raises questions about what the information represents and its relation to other non-feedforward input to visual cortex (Meyer, 2012; Smith, & Muckli, 2010). Here, we use high-resolution 7T functional brain imaging and multivariate classifiers to study auditory information and mental imagery in the early visual cortex of blindfolded subjects. **Methods** Five blindfolded subjects took part in a 7T fMRI experiment. Subjects either listened to one of three sounds (forest, traffic, people) or were cued to engage in mental imagery of one of the scenes for 12 seconds. We used polar angle and eccentricity retinotopic mapping to localise V1, V2 and V3. We ran a multivariate searchlight analysis (Kriegeskorte, Goebel, & Bandettini, 2006) in which the activity pattern from a cluster of voxels centred on every voxel in the early visual cortex was entered into a support vector machine classifier. Thus, each voxel was labelled with decoding accuracy based on nearby information. The information profile across entire V1 cortical depths was also analysed. **Results** Results suggest that both auditory and imagery information can be classified from non-stimulated visual cortex. There seems to be some commonality in the location of the patches of early visual cortex that can classify auditory and imagery information. While several possibilities remain open at this stage, there is a hint that during auditory stimulation information seems to be localised more to deeper layers of V1 (see figure 1). **Conclusions** Our data suggest that feedback to early visual cortex occurs - even in the absence of feedforward stimulation. Moreover, non-visual sensory input reaches the visual cortex at the earliest levels and seems to be localized in a similar way as feedback based on mental imagery.



Disclosures: **M. Bennett:** None. **L.S. Petro:** None. **A.A. Morgan:** None. **F. De Martino:** None. **L. Muckli:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.24/JJ11

Topic: D.07. Vision

Support: NSF GRFP DGE-114747

1RO1EY02231801A1

NRSA 1F31EY027201-01

Title: Development differentially sculpts receptive fields across human visual cortex

Authors: *J. GOMEZ[1], V. S. NATU[2], B. L. JESKA[2], M. A. BARNETT[3], K. GRILL-SPECTOR[4]
[2]Psychology, [1]Stanford Univ., Stanford, CA; [3]Univ. of Pennsylvania, Philadelphia, PA; [4]Psychology, Stanford Univiersity, Stanford, CA

Abstract: A fundamental property of neurons in the visual system is that they have receptive fields that process visual information in spatially restricted regions of visual space. How RFs develop in humans, however, is unknown. While high-level visual areas involved in reading and face recognition show a protracted development in humans, it is unknown if this development is associated with changes to receptive fields, and if so, at what stages of the visual hierarchy they occur. To answer these questions, we estimate the population receptive field (pRF) of all voxels in the visual system in 26 typical children (5-12 years) and 26 adults (22-27 years) and compared across age groups. Participants took part in two fMRI experiments: (1) retinotopic mapping with bars containing black and white checkerboards to delineate retinotopic areas and estimate pRFs, and (2) a functional localizer containing faces, words, bodies, places, and objects to map high-level regions. In the ventral stream, we find no significant difference across age groups in polar angle, eccentricity, pRF size, or visual field coverage in areas V1-VO1 (Figure 1A). In contrast, we found differential development of pRFs of face- and word-selective regions across hemispheres. In the right hemisphere, pRFs and coverage in face-selective cortex become more foveally biased from childhood to adulthood, while word-selective cortex becomes less so. In the left hemisphere, this pattern is reversed (Figure 1B). We link this pRF development to behavior, collecting measurements of fixation patterns on faces and words outside the scanner. We find an increased foveal bias on words and faces in adult relative to child participants (Figure 1C). These results thus suggest a differential development of pRF and visual field coverage in face and word selective regions across hemispheres, which may importantly be driven by changes in viewing experience across development.

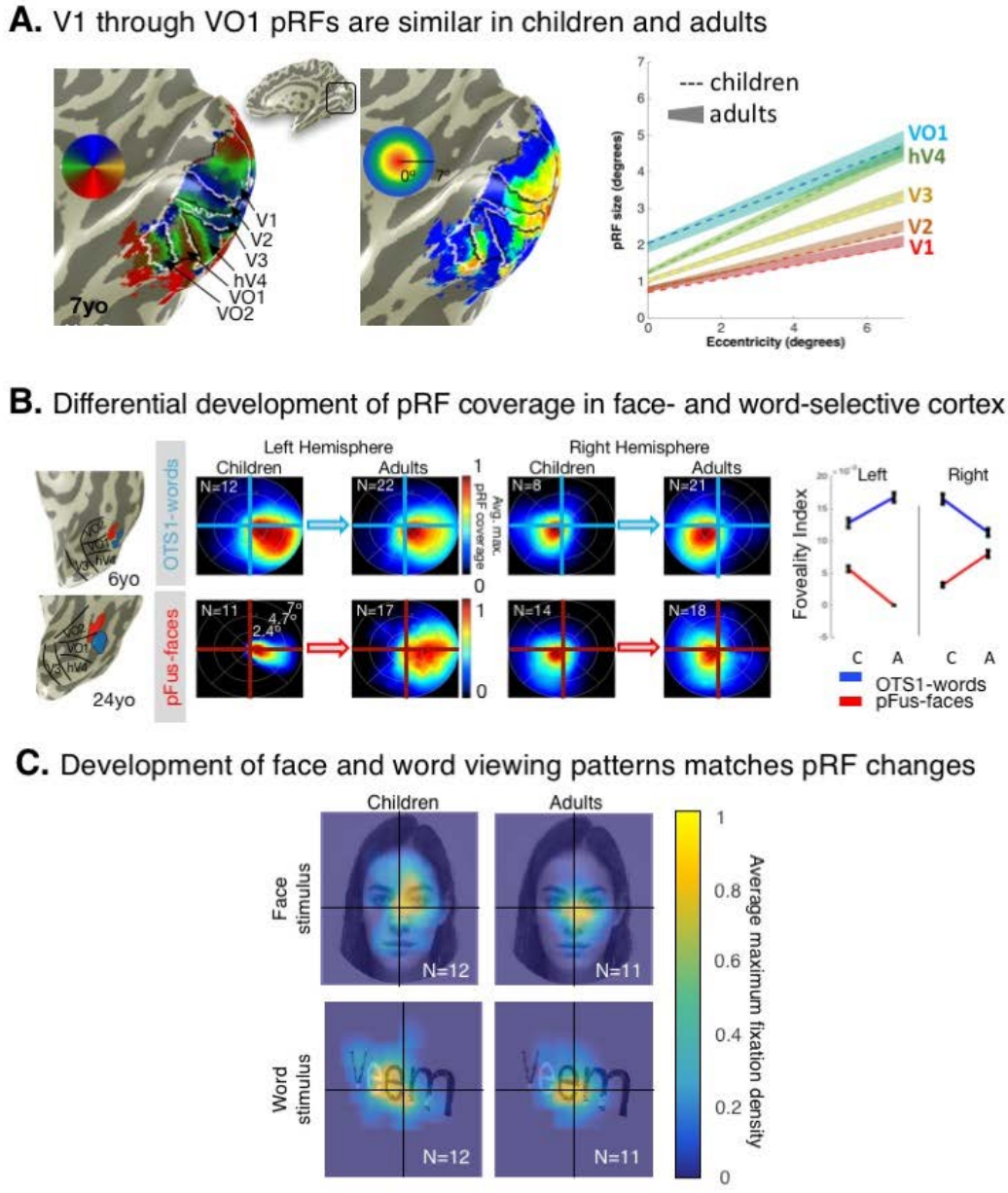


Figure 1. (A) Example pRF phase and eccentricity fits on the cortically inflated surface of a 7 year old child. On the right, average pRF eccentricity vs. size relationships across all children (dotted lines) and adults (shaded standard error of the mean). **(B)** Visual field coverage of face- and word-selective cortex (example fROIs shown on left) averaged across subjects. Foveality index calculated by multiplying coverage map by a mexican-hat filter positively weighting foveal coverage. C=children, A=adults. **(C)** Eye tracking experiment performed outside scanner in which fixations on stimuli are smoothed and averaged across subjects. Fixations are more central and punctate in adults.

Disclosures: J. Gomez: None. **V.S. Natu:** None. **B.L. Jeska:** None. **M.A. Barnett:** None. **K. Grill-Spector:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.25/JJ12

Topic: D.07. Vision

Support: NWO/CAS 012.200.012

ABMP VU/UvA ATT #2

Title: Retinotopic organization in the default mode network

Authors: *T. KNAPEN[1], D. VAN ES[2], M. BARENDREGT[2]
[1]VU Amsterdam, Amsterdam, Netherlands; [2]Cognitive Psychology, Vrije Univ. Amsterdam, Amsterdam, Netherlands

Abstract: The brain's default mode network (DMN) comprises a set of brain regions that consistently show decreases in Blood Oxygenation Level Dependent (BOLD) signal during task engagement, at times when most other cortical areas show BOLD signal increases. Despite being a well-known property of the DMN, the nature and function of DMN deactivations remains unclear.
Here we demonstrate that a fundamental neuroanatomical principle, retinotopy, exists within the human DMN. Specifically, BOLD decreases in the lateral parietal node of the DMN are specific to the appearance of a visual stimulus in a circumscribed region of retinotopic space. We estimated population receptive fields (pRFs) of negative amplitude from BOLD time courses in the DMN, and show that this region contains a coherent retinotopic map along the cortical surface. Moreover, this description of spatial preferences in the DMN, combined with ongoing activation patterns, allows us to reconstruct (decode) the position of a visual stimulus with a fidelity comparable to the known retinotopic maps of the intraparietal sulcus.
Our results indicate that retinotopically specific activations and deactivations synergistically subserve the processing of visual information. As DMN regions have been shown to contain pRFs that selectively activate for social information, it is likely that representations in retinotopic and other reference frames coincide in these regions. This overlap would allow local computations to integrate multiple levels of information processing.

Disclosures: **T. Knapen:** None. **D. van Es:** None. **M. Barendregt:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.26/JJ13

Topic: E.07. Rhythmic Motor Pattern Generation

Support: Canadian Institutes of Health Research (CIHR)

German Research Foundation (DFG) CRC 889, project C9

Title: Separate resting state networks for grasping and visually guided reaching in macaques

Authors: *R. S. GREULICH[1], R. ADAM[2,3], S. EVERLING[2,4], H. SCHERBERGER[1,5]
[1]German Primate Ctr., Goettingen, Germany; [2]Robarts Res. Inst., [3]Grad. Program in Neurosci., [4]Physiol. and Pharmacol., Univ. of Western Ontario, London, ON, Canada; [5]Fac. of Biol. and Psychology, Univ. of Göttingen, Göttingen, Germany

Abstract: Grasping allows primates to handle food and tools in a very specialised way. Tract tracing studies in nonhuman primates have highlighted three cortical areas as central nodes in the grasping network: anterior intraparietal area (AIP), ventral premotor area (F5), and the hand area of primary motor cortex (M1) (Davare et al., 2011). Although the anatomical connectivity between these cortical areas is known, their functional connectivity is less well understood. Furthermore, neurons were found which responded to grasping in the visual area V6a and the medial intraparietal area (MIP) (Galletti et al. 2003, Gamberini et al. 2011). Area V6a and MIP are part of the dorsal stream and are involved in the processing of visually guided reaching movements (Baccalá and Sameshima, 2001).
Here, we aim to expand our understanding of the functional connectivity within and between the cortical grasping and reaching networks in macaques using resting-state functional MRI. We applied a seed based functional connectivity analysis for each region-of-interest individually (AIP, F5, M1, V6a, MIP) and compared the resulting correlation maps.
First, we found that AIP, F5, and M1 individually reveal similar resting-state networks. These areas are part of the grasping network, which extends over large parts of the parietal lobe and includes somatosensory and motor areas. Area V6a and MIP also revealed comparable resting-state networks, which covered posterior frontal areas around the frontal eye field and extending to the temporal lobe.
These findings reveal two distinct networks, one traditionally attributed for grasping and one for visually guided reaching, with only slight overlap between the two.

Disclosures: R.S. Greulich: None. **R. Adam:** None. **S. Everling:** None. **H. Scherberger:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.27/JJ14

Topic: D.07. Vision

Support: NIH Grant 1R01EY02391501A1

DFG Grant GR 4850/1-1

Title: A preference for mathematical processing outweighs selectivity for Arabic numbers in the inferior temporal gyrus

Authors: *M. GROTHEER[1], B. L. JESKA[1], K. GRILL-SPECTOR[1,2,3]
[1]Dept. of Psychology, [2]Stanford Neurosciences Inst., Stanford Univ., Stanford, CA;
[3]Neurosciences Program, Stanford Univ. Sch. of Med., Stanford, CA

Abstract: A region in the inferior temporal gyrus (ITG), referred to as the 'number form area' has been shown to respond more strongly to Arabic numbers than other visual stimuli, suggesting that it is involved in the visual encoding of numbers. However, other studies report that this region activates during mathematical tasks on auditory stimuli, suggesting that it may be involved in mathematical rather than visual processing. To address this debate, we conducted three fMRI experiments (N=15), in which we varied stimuli and task to determine how these factors affect ITG responses. In Experiment (Exp) 1, we measured responses to visually different stimuli (numbers, letters, false numbers and letters, Fourier scrambled numbers and letters, and objects), during a non-mathematical task (1-back on stimulus identity). In Exp 2, we measured responses to well-controlled character-like stimuli (colored number and letter morphs) during different tasks (adding, reading, and remembering stimulus color). In Exp 3, we measured responses to visually dissimilar stimuli (colored numbers, hands, and dice) during different tasks (adding and remembering color). Results show that those voxels in the ITG showing a preference for numbers vs. other visual stimuli in Exp 1, show higher responses to mathematical processing (adding vs. color or reading) in Exps 2 and 3, but not higher responses to numbers vs. other stimuli in Exps 2 or 3. Likewise, those voxels in the ITG showing higher responses to mathematical processing (adding vs. color or reading) in Exp 2, also show higher responses to mathematical processing (adding vs. color) in Exp 3 using different stimuli, but do not show a preference for numbers in any of the experiments. Finally, a classifier approach revealed that mathematical task can be successfully decoded (>85% accuracy in both Exps 2 and 3) from distributed ITG responses, but decoding accuracy for numbers was poor (<65% in Exp 1, at chance in Exp 2 and <45% in Exp 3). Together, these results suggest a need to rethink the function of the "number form area" as the context of a mathematical task rather than the processing of number stimuli produces reliable local and distributed ITG activations. We

propose that the ITG ascribes numerical content to the input, irrespective of the nature of the stimulus.

Disclosures: M. Grotheer: None. **B.L. Jeska:** None. **K. Grill-Spector:** None.

Poster

403. Visual Cortical Streams: Primate and Human

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 403.28/JJ15

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Title: A neural basis for biased competition in the developing visual system: An fMRI study in school-aged children

Authors: *N. KIM, M. A. PINSK, S. KASTNER
Princeton Univ., Princeton, NJ

Abstract: Attention functions develop significantly in childhood. Prior research investigated age-related differences in performance on various attention tasks and found different developmental trajectories for exogenous and endogenous attention and for sustained, selective, and executive attention. However, neural mechanisms underlying attention functions during development remain unclear. Here, we used hypotheses derived from biased competition model (Desimone and Duncan, 1995) to explore the neural basis of visual attention in elementary school children. An important role of attention is to select or ignore components of sensory input to optimize behavior. According to the biased competition model, multiple stimuli that are present simultaneously compete for neural representation due to the limited processing capacity of the visual system. The neural competition can be influenced in several ways including bottom-up and top-down factors. To examine sensory competition in the developing visual system, we presented multiple color patches either simultaneously or sequentially while children (age 6 to 12) were engaged in an irrelevant fixation task. We hypothesized that, as previously shown in the adult brain, multiple stimuli would compete with one another when presented simultaneously and thus mutually suppress neural responses in multiple areas of the visual processing hierarchy. We quantitatively evaluated the degree of sensory competition in multiple topographically organized areas, as identified with the help of a probabilistic functional brain atlas. Our results on sensory competition will provide a basis for investigating how top-down and bottom-up factors influence neural competition, thereby providing a neural basis for the filtering of unwanted information during the selection process. The results of these studies may have implications for a better understanding of neurodevelopmental disorders such as attention deficit disorder.

Disclosures: N. Kim: None. **M.A. Pinsk:** None. **S. Kastner:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.01/JJ16

Topic: D.08. Vestibular System

Support: NIH/NIDCD grant DC013798 (SMR)

DC008846 (GRH)

Title: Optical infrared vestibular stimulation evokes physiological eye movement and cardiovascular responses

Authors: *W. JIANG[1], D. RICE[3], G. P. MARTINELLI[4], G. R. HOLSTEIN[5], S. RAJGURU[2]
[1]Biomed. Engin., Univ. of Miami, Miami, FL; [2]Biomed. Engin. and Otolaryngology, Univ. of Miami, Coral Gables, FL; [3]Univ. of miami, Miami, FL; [4]Dept. Neurol., [5]Depts Neurol, Neurosci, Anat/Cell Bio, Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Infrared neural stimulation has been demonstrated to modulate the pattern of neural signals transmitted from the angular motion sensing semicircular canals of the vestibular system to the brain. The goal of the present study was to investigate the physiological effects of pulsed infrared stimulation (pIR) focused on individual vestibular end organs in the rat. A surgical approach was developed to target specific semicircular canals and otolith organs and pIR (1863nm, 250pps, 200µs) was directed to vestibular endorgans using custom-designed 200 and 400 µm optical fibers. The evoked bilateral eye movements were measured using a video-based, two-channel eye tracking system (ISCAN Inc, Woburn, MA) and changes in blood pressure and heart rate were recorded as a measure of activation of the vestibular sympathetic reflex pathway. The activation of vestibulo-ocular motor pathways by frequency modulated pIR evoked significant characteristic bilateral eye movements. The eye movements continued through 30+ minutes of continuous pIR stimulation. Simultaneously, a significant drop in blood pressure and heart rate were recorded with low frequency sinusoidal pIR stimulation. The results compared well with our previous observations that sinusoidal galvanic vestibular stimulation and tilt can be used to activate central vestibular neurons of the vestibulo-sympathetic reflex pathway. To identify the vestibular endorgans activated by pIR, histology and micro-computed tomography were performed. Following the experiments, the radiation energy was increased to a level likely to induce tissue damage. The cochlea and vestibular end organs were harvested to visualize the damage and determine the beam path. In other animals, high-resolution microCT was used to image the inner ear structures and the location of the optical fiber fixed in place following experimentation. Overall, the results demonstrate selective stimulation of vestibular endorgans by pulsed infrared optical stimuli and suggest potential applications in inner ear.

Disclosures: **W. Jiang:** None. **D. Rice:** None. **G.P. Martinelli:** None. **G.R. Holstein:** None. **S. Rajguru:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.02/JJ17

Topic: D.08. Vestibular System

Support: NIH/NIDCD Grant DC008846 (GRH)

NIH/NIDCD Grant DC013798 (SMR)

Title: Localization of central vestibular neurons activated by pulsed infrared stimulation of individual vestibular end organs

Authors: ***G. R. HOLSTEIN**[1], D. RICE[3], W. JIANG[3], S. RAJGURU[4], G. P. MARTINELLI[2]
[1]Depts Neurol, Neurosci, Anat/Cell Bio, [2]Dept. Neurol., Icahn Sch. of Med. at Mount Sinai, New York, NY; [3]Biomed. Engin., Univ. of Miami, Miami, FL; [4]Biomed. Engin. and Otolaryngology, Univ. of Miami, Coral Gables, FL

Abstract: The vestibulo-sympathetic reflex (VSR) pathway is important for modulating blood pressure upon changes in head position with regard to gravity, as occurs when humans rise from a supine position and when quadrupeds climb or rear. We have demonstrated that sinusoidal galvanic vestibular stimulation (sGVS) and tilt can be used to activate central vestibular neurons of the VSR pathway. Changes in blood pressure were recorded in response to these stimuli, and the activated vestibular neurons were identified by cFos labeling and retrograde tract tracing. Maps of the activated VSR neurons indicate that the cells are located almost exclusively in parvocellular medial vestibular nucleus (MVN), the caudal region of MVN, and throughout the caudal half of the spinal vestibular nucleus (SpVN). However, both sGVS and tilt activate the vestibular nerve globally.
The goal of the present study was to identify the specific locations and densities of VSR neurons activated by stimulation of individual vestibular end organs in rats. To achieve this, pulsed infrared laser stimulation (pIRs) at 1863nm, 100-250pps, and 250µs was directed through the round window toward vestibular end organs in rats using customized optical fibers. Changes in BP and heart rate and evoked eye movements were recorded in response to pIRs of semicircular canal cristae and otolith maculae. Cells that were activated by pIRs were identified by cFos/DAB immunohistochemistry. Labeled cells in the vestibular nuclei were counted in skip-serial sections through the caudal vestibular nuclei that were separated by at least 100 µm. The cell counts from each rat were mapped onto 16 representative rostro-caudal Bregma levels. Results indicate that posterior canal stimulation activates primarily (but not exclusively) ipsilateral neurons in both

parvocellular MVN and SpVN. Although VSR neurons were observed between Bregma -11.16 and -12.84, the highest density of VSR neurons activated by posterior canal stimulation was concentrated at Bregma levels between -11.52 and -12.12. This distribution is contrasted with the maps of activated VSR neurons resulting from global vestibular nerve stimulation, and from pIRs of individual otolith organs.

Disclosures: G.R. Holstein: None. **D. Rice:** None. **W. Jiang:** None. **S. Rajguru:** None. **G.P. Martinelli:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.03/JJ18

Topic: D.08. Vestibular System

Title: Probing the role of the efferent vestibular system using direct monosynaptic viral tracing

Authors: M. A. MATHEWS[1], *A. J. CAMP[2], A. J. MURRAY[3]
[1]Univ. of Sydney, Sydney, Australia; [2]Univ. of Sydney, Sydney University, Australia; [3]Sainsbury Wellcome Ctr. for Neural Circuits and Behaviour, Univ. Col. London, London, United Kingdom

Abstract: *Background:* Many sensory modalities adopt efferent circuitry in the central nervous system (CNS) to modulate peripheral targets. Unlike other sensory modalities, the functional role of the efferent vestibular system (EVS), originating in the brainstem efferent vestibular nucleus (EVN) and terminating on vestibular hair cells and afferent fibres at the periphery, remains elusive. Previous work has focused on the anatomy, morphology, physiology, synaptic mechanisms, and peripheral responses to EVN activation, with few studies directly investigating a behavioural function. Moreover, a lack of understanding regarding EVN circuit dynamics makes it impossible to link vestibular efferent activity to behaviour. *Objective:* Here, we sought to determine the central context within which the EVN is activated by tracing the monosynaptic inputs to this group of neurons, and expand on the work presented at the previous Neuroscience meeting. *Methods:* We used monosynaptic rabies tracing to determine the direct inputs to EVN neurons. In mice (n =3) expressing Cre under the control of the choline acetyl transferase (ChAT) promotor we expressed the rabies glycoprotein (G) selectively in EVN neurons. Following G expression, we injected glycoprotein-deficient rabies (RABVG) virus expressing a fluorescent protein into the horizontal and posterior semicircular canals in the inner ear of the same animal. Histological analysis was performed to identify the direct inputs to EVN neurons. *Results:* We observed >50 direct inputs from diverse regions throughout the brainstem, cerebellum, and forebrain including from structures in the telencephalon and diencephalon. *Conclusions:* The identification of direct monosynaptic inputs to mouse EVN neurons with

rabies virus will allow us to expand hypothesis regarding EVN function. In addition, this method will allow us to manipulate EVN neurons, or their inputs, via electrical, chemical or optogenetic methods providing a means to systematically explore the context-dependent central modulation of peripheral vestibular function.

Disclosures: M.A. Mathews: None. **A.J. Camp:** None. **A.J. Murray:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.04/JJ19

Topic: D.08. Vestibular System

Support: NSERC Discovery Grant

CIHR

CIHR doctoral fellowship to C.M.

Title: Decoding body-centered representations of translational motion in 3D from neural activity in the rostral fastigial nucleus

Authors: *C. MARTIN, J. X. BROOKS, A. M. GREEN
Neurosciences, Univ. De Montreal, Montreal, QC, Canada

Abstract: Many of our daily behaviors (e.g., postural control, locomotion, reaching) rely on estimates of our body motion. Vestibular signals are essential for such estimates but to contribute appropriately they must be transformed from a head- to a body-centered reference frame. Recently, we extended previous work to examine whether cells in the rostral fastigial nucleus (rFN) reflect this transformation fully in 3D. Consistent with such a transformation, many cells exhibited spatial tuning shifts for changes in head-re-body orientation in both horizontal and vertical planes. However, tuning properties were broadly distributed between head- and body-centered with most cells reflecting only a partial transformation. This suggests that despite evidence in the rFN for the precise matching of vestibular and neck proprioceptive signals required to distinguish head from body movement (Brooks and Cullen, 2009) explicit estimates of body motion are unlikely to be encoded by most individual cells. The goal of the current study was to investigate the facility with which fully body-centered representations in 3D could be decoded from populations of rFN cells and whether specific tuning properties affected this capacity. To this end, we generated a synthetic population of 500 cells with preferred tuning spanning 3D space and having distributions of spatio-temporal tuning properties similar to those of our recorded neurons. We then examined the ability to decode fully body-centered motion representations along 13 axes in 3D space across changes in head-re-body orientation of ± 90° in

the yaw, pitch and roll planes. We show that fully body-centered representations can be decoded through a simple linear combination of the tuning functions of small groups (5-7) of these cells. Furthermore, with changes in decoding weights such representations can exhibit a broad range of temporal properties ranging from lagging translational velocity to leading acceleration. Two key properties of rFN cells were crucial to this capacity. These include: 1) a near absence of "gain-fields"; 2) tuning shifts that follow a very specific theoretically predicted trajectory in 3D space. Collectively, these observations suggest that despite the broad distribution of tuning properties, rFN cells reflect a late or "output" stage in the head-to-body reference frame transformation. We propose that the maintenance of partially transformed responses with different spatio-temporal properties facilitates the creation of downstream body motion representations with a broad range of dynamic characteristics, consistent with the functional requirements for tasks such as postural control.

Disclosures: C. Martin: None. **J.X. Brooks:** None. **A.M. Green:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.05/JJ20

Topic: D.08. Vestibular System

Support: Canadian Institutes of Health Research

Title: Noise correlations between neurons in early vestibular pathways are negligible during both active and passive self-motion

Authors: *A. DALE[1], J. CARRIOT[2], M. J. CHACRON[2], K. E. CULLEN[2,3]
[2]Physiol., [1]McGill Univ., Montreal, QC, Canada; [3]Biomed. Engin., Johns Hopkins Univ., Baltimore, MD

Abstract: Vestibular sensory information is encoded by the primary afferents of the VIIIth cranial nerve and transmitted to the brainstem vestibular nuclei. To fully understand how self-motion signals are decoded by central processing stages, correlated variability (i.e., noise correlations) among ensembles of neurons must be taken into account (e.g., Cohen and Kohn 2011). Notably, with shared afferent input among nearby vestibular nuclei neurons (Sato et al. 1989), as well as evidence of gap junctions between them (Condorelli et al. 2000), one might predict that noise correlations play a role in early vestibular processing during everyday activities. However, to date, this question remains open. Here, we computed cross correlations between the activity of pairs of vestibular nuclei neurons as well as their afferent inputs during 1) sinusoidal passive stimulation at frequencies from 0.5-4Hz, 2) passive stimulation with active-like trajectories, and 3) active movements. We first show that the resting discharges of both

peripheral afferent and central vestibular only neurons exhibit negligible noise correlations over windows ranging from 1-50ms. We next considered the effect of self-motion stimuli resulting from different behavioral contexts. We found that neuronal activities displayed insignificant noise correlations during passive sinusoidal stimulation which maximally drives vestibular sensory neurons. Furthermore, noise correlations remained negligible during more natural passive vestibular stimulation. Finally, we tested vestibular only neurons during voluntary self-motion when their responses are cancelled by purported cerebellar inputs to the vestibular nuclei. We found that over durations spanning synchronous firing to whole movements, the responses of vestibular nuclei neurons remained independent from one another. As such, we conclude that if anatomical connections tend to correlate activity between neurons, then additional factors must be present to cause trial-to-trial variabilities of neuronal responses to be de-correlated. We hypothesize that this independence between neuronal responses allows subsequent stages of vestibular processing (e.g., thalamus or cortex) to best average out the variability in their inputs in order to achieve improved stimulus coding.

Disclosures: **A. Dale:** None. **J. Carriot:** None. **M.J. Chacron:** None. **K.E. Cullen:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.06/JJ21

Topic: D.08. Vestibular System

Support: ANR-12-BSV4-0027

ANR-15- CE37-0007

ANR-10-LABX-54 MEMO LIFE

ANR-11-IDEX-0001-02 PSL

Title: Cerebellar re-encoding of self-generated head movements

Authors: ***G. P. DUGUÉ**[1], M. TIHY[1], B. GOURÉVITCH[2], C. LENA[3]
[1]IBENS, Paris, France; [2]Dept. Cognition & Behavior, Inst. De Neurosci. Paris-Saclay (neuropsi), Orsay, France; [3]Inst. de Biologie de l'Ecole Normale Superieure, PARIS, France

Abstract: Head movements are primarily sensed in a reference frame tied to the head, yet they are used to calculate self-orientation relative to the world. This requires to re-encode head kinematic signals into a reference frame anchored to earth-bound landmarks such as gravity, through computations whose neuronal substrates remain to be determined. Here, we studied the encoding of self-generated head movements in the caudal cerebellar vermis, an area essential for

graviceptive functions. Multi-unit recordings were obtained from caudal cerebellar units in freely moving rats, while head kinematics was monitored using a miniature digital inertial sensor. This sensor provided the same type of information as the animal's vestibular organs (a measurement of the instantaneous head angular velocity and acceleration).The gravitational component of acceleration was isolated offline using an orientation filter algorithm. The kinematics of natural head movements in our conditions was characterized by 1) a multi-peaked power spectrum of angular velocity signals up to 20 Hz, 2) a predominance of the effect of gravity in the lower frequency range of acceleration (< 2 Hz) and 3) the relative absence of translational acceleration in the total acceleration signal (i.e. most of the non-gravitational acceleration signal was generated by rotations). We found that, contrarily to peripheral vestibular inputs, most caudal Purkinje cells exhibited a mixed sensitivity to head rotational and gravitational information (with a negligible contribution of non-gravitational acceleration information), and were differentially modulated by active and passive whole-body movements. The presence or absence of visual cues did not seem to condition the units' responses to head movements. We analyzed in greater details the units' receptive fields and found that they were tuned to a preferred head rotation whose direction depended on head tilt. In a subpopulation of cells, this tilt dependent rotation-coding scheme underlay a tuning to angular velocity about an axis defined relative to gravity. These cells thus exhibit a transformed angular velocity signal which is mapped to an external, earth-bound reference frame, and could provide the appropriate substrate for computing gravitationally-anchored head direction signals.

Disclosures: **G.P. Dugué:** None. **M. Tihy:** None. **B. Gourévitch:** None. **C. Lena:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.07/JJ22

Topic: D.08. Vestibular System

Support: CIHR

Title: Temporal whitening of naturalistic self-motion stimuli by early vestibular pathways

Authors: *D. E. MITCHELL[1], A. KWAN[2], J. CARRIOT[3], M. J. CHACRON[2], K. E. CULLEN[2]
[1]Dept. of Physiol., [3]Physiol., [2]McGill Univ., Montreal, QC, Canada

Abstract: Understanding how the brain generates appropriate behavior and accurate perception in response to incoming sensory signals remains a fundamental challenge in neuroscience. The common wisdom is that neural coding strategies are adapted to the statistics of the sensory signals found in the natural environment. Specifically, early sensory pathways are thought to

efficiently process natural stimuli by matching both their tuning properties and neural variability to the statistics of their natural stimuli, thereby removing redundancy and thus maximizing information transmission. If this is the case the resulting neural response is independent of frequency (i.e. “whitened”). To date, however, there has been no experimental demonstration that such a match exists. Here we show for the first time how whitening is achieved by sensory pathways. Specifically, we examined how neuronal variability and tuning are balanced to optimize coding of naturalistic self-motion in early vestibular pathways. We recorded from vestibular neurons during naturalistic self-motion and found that whitening occurs sequentially, beginning at the level of the sensory periphery, and is then further refined centrally. We found that, while the tuning of afferents was largely sufficient to explain their contribution to whitening, this was not the case for central neurons. Indeed, the exceptionally whitened responses of central neurons could only be accounted for when we considered their resting discharge variability as well as their high-pass tuning. Our analysis then further revealed that this combination of resting power and variability is well matched to offset naturalistic stimulus statistics in order to temporally whiten neural responses, thereby maximizing information transmission. These findings overturn the prevailing view that whitening of neural responses is achieved through tuning alone by demonstrating that neural variability is a fundamental feature of the neural code.

Disclosures: D.E. Mitchell: None. **A. Kwan:** None. **J. Carriot:** None. **M.J. Chacron:** None. **K.E. Cullen:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.08/JJ23

Topic: D.08. Vestibular System

Support: GRF HKU761812M

GRF HKU762313M

GRF 17131816

HKU Strategic Research Theme on Neuroscience

Title: Neonatal excitation-inhibition imbalance introduces long-lasting changes to neuronal recruitment in vestibular circuits for spatial navigation

Authors: *O. W. CHUA, Q. F. JIANG, K. L. K. WU, Y. S. CHAN
Sch. of Biomed. Sci., The Univ. of Hong Kong, Hong Kong, China

Abstract: A critical period exists during the first 2 postnatal weeks for development of GABAergic transmission in the rat vestibular nucleus (VN). Perturbation of GABAergic transmission in the VN with bicuculline, a $GABA_A$ blocker, during this period abolished efficient spatial navigation behaviour in adults. Post-critical period treatment at P21 had no effect. We hypothesized that the behavioural deficits are the result of long-lasting changes in MVN circuitry. The number of parvalbumin-expressing neurons and perineuronal nets was increased in rats treated with bicuculline at P1 as compared with controls. We further found that there was a decrease in amplitude/frequency of sIPSCs and change in firing pattern of projecting vestibular neurons in adult rats with bicuculline treatment at P1. Since the anterodorsal thalamic nucleus (ADN) integrates vestibular input to generate head direction signal for navigation, we examined neuronal activation in the ADN in these rats after natural vestibular stimulation. It was found that *c-fos* expression pattern in the ADN of P28 rats after wobble rotation was deranged. Taken together, the data show that neonatal disruption of excitation/inhibition balance in the VN leads to long-lasting derangement of MVN circuitry and its output thereby impacting vestibulo-thalamic neuron recruitment in rats and navigational behaviour. Our findings will inform strategies to promote rewiring of brain circuits for rehabilitation.

Disclosures: O.W. Chua: None. **Q.F. Jiang:** None. **K.L.K. Wu:** None. **Y.S. Chan:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.09/JJ24

Topic: D.08. Vestibular System

Support: University of Debrecen, Faculty of Dentistry

NKFIH-K 115471

MTA-TKI 11008

Title: Vestibular compensation: possible role of extracellular matrix

Authors: *K. MATESZ[1,2]
[1]Univ. of Debrecen, Fac. of Dent., Debrecen, Hungary; [2]Dept. of Anatomy, Histology and Embryology, Oral Anat. Div., Univ. of Debrecen, Debrecen, Hungary

Abstract: Peripheral lesion of the vestibular system (VS) evokes postural and visual deficits accompanied by impaired vegetative function. Depending on the species, most of the symptoms regain spontaneously during the process of vestibular compensation (VC). In amphibians, the sectioned vestibular nerve regenerates, the re-growing axons establish functional synapses. In contrast, the injured vestibular nerve of mammalian species does not regenerate but functional

recovery from the vestibular symptoms is possible. Over the last few decades, the role of extracellular matrix (ECM) on the plasticity and regeneration of CNS has been emphasized. Our hypothesis is that different molecular composition of ECM in the perineuronal nets (PNNs) of vestibular neurons, the condensed ECM around the neuronal cell bodies and dendrites, is responsible for the different restoration capacity of VS in non-mammalian and mammalian species. Therefore, we examined the expression of ECM during the VC in the PNN of the vestibular nuclei in frog and rat. On anesthetized frogs the vestibular nerve was transected and reunited, in the rats unilateral labyrinthectomy was performed to destroy the vestibular sense organs. The animals were sacrificed at different survival times and on the brainstem section the expression of hyaluronan (HA), chondroitin sulfate proteoglycans (CSPGs), tenascin-R and link proteins were detected by using histochemical and immunohistochemical reactions. In the non-operated frog and rat conspicuous molecular differences were shown in the PNNs. During the VC, the time course of bilateral destruction and subsequent reorganization of PNNs showed species differences and corresponded to the restoration of static symptoms. Our results suggest that the permissive HA-rich, but CSPG-free PNNs of frog vestibular nuclei may be associated with the high degree of plasticity and regenerative potential of VS in amphibians. In contrast, the strong expression of the CSPGs in the PNNs of the rat vestibular nuclei seems to be against the plasticity of VS in mammalian species. This contradiction can be resolved by the very quick and dynamic quantitative and qualitative changes in the expression of ECM molecules in the rat PNNs presented in our experiments. It is tempting to assume that in the non-lesioned rat the condensed PNNs are needed to stabilize the synaptic contacts, whereas their postlesional disruption has beneficial effect on the synaptic reorganization in order to compensate the lost vestibular input. On the other hand, the loose ECM network in the PNN may allow the in-growth of non-vestibular axon terminals as a possible factor of VC.

Disclosures: K. Matesz: None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.10/JJ25

Topic: D.08. Vestibular System

Support: GWU Dept Anatomy Research Funds

GWU Luther -Rice Undergraduate Fellowship

Title: Malformation of the vestibular inner ear leads to abnormal vestibular nuclei development

Authors: S. J. LILIAN[1], H. E. SEAL[1], A. POPRATILOFF[1], J. C. HIRSCH[1], *K. D. PEUSNER[2]

[1]Anat. and Regenerative Biol., [2]George Washington Univ. Sch. of Med. and Hlth. Sci., Washington, DC

Abstract: Many developmental disorders affecting the inner ear are manifested clinically as delayed motor development and challenges in maintaining posture and balance, indicating that central vestibular circuits are affected. How central vestibular wiring is changed in pediatric cases with inner ear abnormalities is poorly understood. We hypothesize that the malformed vestibular inner ear transmits abnormal signals to developing vestibular nuclei, the first brain centers processing the input. We expect abnormal signals to influence the development of the entire vestibular neural circuitry from brainstem vestibular nuclei to vestibular cerebral cortex. Our lab has implemented the chick embryo, a well-established animal model for studying normal development, as a novel model to study vestibular brain development in congenital disorders. Existing genetic mutant mouse models show high variability in inner ear pathology, possibly resulting in many variable brain abnormalities. We find that surgical rotation of the developing inner ear, or otocyst, 180 degrees in two-day-old chick embryos results in an abnormal inner ear with one of two forms. The predominant defect is a large sac with truncated or absent canals, while the less frequent pathology is an inner ear with one or more small canals. These defects are similar to those commonly reported in pediatric cases of congenital vestibular inner ear disorders. Thus, the chick offers a reproducible model with phenotypic outcomes like those found in children with congenital vestibular inner ear disorders. After hatching, chicks with rotated otocyst show abnormal vestibular behaviors, including head tilt, stumbling while walking, delayed righting reflex, and a tendency to close one or both eyes.
To understand the consequences of this peripheral manipulation on brainstem vestibular centers, we began by counting vestibular nuclei neurons in the chick tangential nucleus after otocyst rotation. The tangential nucleus was selected because its principal cells are distinctive vestibular second-order projection neurons readily identified in the lateral medulla oblongata. We find significant reduction in the number of neurons in the tangential nucleus on the rotated side. Using the chick model, we will test fundamental neuronal mechanisms operating during the emergence and maturation of vestibular signal processing and compare the results to those reported in normal chicks (Peusner, 2014). Understanding the role of vestibular sensory input on brain development may help to reveal the aberrations in central vestibular neuronal networks, and lead to advances in treating children with malformed vestibular inner ears.

Disclosures: S.J. Lilian: None. **H.E. Seal:** None. **A. Popratiloff:** None. **J.C. Hirsch:** None. **K.D. Peusner:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.11/JJ26

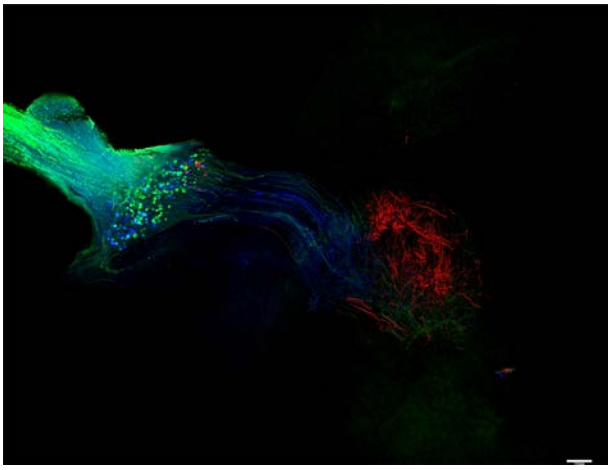
Topic: D.08. Vestibular System

Support: NSERC

Title: Primary projections of the VIIIth nerve in two species of snakes, the western diamondback rattlesnake (Crotalus atrox) and amazon tree boa (Corallus hortulanus)

Authors: *R. M. LONG[1], M. S. BOTHE[2], C. GUTIERREZ-IBANEZ[1], T. KOHL[2], H. LUKSCH[2], H. STRAKA[3], D. R. WYLIE[1]
[1]Neurosci. and Mental Hlth. Inst., Univ. of Alberta, Edmonton, AB, Canada; [2]Tech. Univ. of Munich, Freising, Germany; [3]LMU Munich - Biocenter Martinsried, Planegg, Germany

Abstract: Detection of angular and translational body motion derives from sensory transformations within otolith and semicircular canal organs. Signals are transmitted to the brain by the VIIIth nerve with an anterior branch that carries afferents from the saccule, utricle, anterior and horizontal canals and by a posterior branch that carries afferents from the posterior canal, lagena and various papillar organs. The central projection of individual organs has been described in various vertebrates, however, these studies generally examined only one organ at a time. Here we used an *in vitro* approach with fluorescent tracers to simultaneously trace the respective termination pattern of afferent fibers from each semicircular canal in two species of snakes (*C. atrox, C. hortulanus*). Brains were isolated and each semicircular canal nerve branch was stained separately with Dextran-Tx Red, Dextran Fluorescein, or Neurobiotin 350. Brains were incubated in cold Ringer's solution at 9°C for 48 hrs, fixed, sliced into 50 µm sections and imaged with fluorescence microscopy. Afferents from the different endorgans terminated throughout the rostrocaudal extent of the vestibular complex in rhombomere 1-8, including a cluster of efferent cell bodies ventromedially in r4. Afferents in the posterior branch terminated within the dorsal region of the lateral, medial and descending vestibular nuclei, as well as the tangential and cochlear nuclei. Afferents in the anterior branch terminated in ventral and medial areas of the vestibular nuclei, with a noticeable differential termination pattern of horizontal and anterior canal afferents. Dense terminal labelling from the horizontal and anterior canals was seen, respectively, in the ventrolateral and medial areas of the vestibular nuclei. In contrast, only few afferents terminated as mossy fibers within the cerebellum. The latter derived from the two vertical but not the horizontal canal. The overall pattern of the vestibular afferent projections in snakes is similar to that of other vertebrates suggesting an evolutionarily conserved central topography.



Disclosures: **R.M. Long:** None. **M.S. Bothe:** None. **C. Gutierrez-Ibanez:** None. **T. Kohl:** None. **H. Luksch:** None. **H. Straka:** None. **D.R. Wylie:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.12/JJ27

Topic: D.08. Vestibular System

Support: NIH Grant F32-DC015157

NIH Grant K08-DC013571

Title: Vestibular nucleus and reticular formation neurons with converging limb and labyrinthine inputs differentially discriminate vestibular signals

Authors: *D. M. MILLER, C. D. BALABAN, A. A. MCCALL
Otolaryngology, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: The CNS processes multiple inputs, including from the vestibular periphery and peripheral proprioceptors, to effect balance control. Neurons that receive vestibular inputs in the vestibular nuclei (VN) and pontomedullary reticular formation (pmRF) have recently been shown to respond to convergent proprioceptive inputs from the hindlimb in conscious cats. However, it is presently unknown whether dynamic vestibular and limb proprioceptive signals are processed differentially in either of these two areas. We have modeled the firing of VN and pmRF neurons during vestibular stimulation and hindlimb movement and determined if vestibular or limb movement signals could be used to predict the histologic location of a given neuron. Single-unit recordings were obtained in conscious cats from neurons in the caudal VN (n=77 neurons) and the pmRF (n=60 neurons). Sinusoidal whole body rotation in the roll plane

was used as the search stimulus. Neurons modulating with the search stimulus were subsequently recorded during 10° ramp-and-hold roll body rotation, 10° ramp-and-hold hindlimb movement and to both stimuli delivered simultaneously. Composite response histograms were fit by an additive model of low and high pass filtered limb and body position signals using least squares non-linear regression. The R^2 values for goodness of fit for the VN and pmRF additive models were 0.91 ± 0.03 and 0.65 ± 0.17, respectively. Forward stepwise logistic regression (Wald criterion) was used to estimate coefficients in order to categorize neurons as belonging to either the pmRF or the VN using the set of parameters from the additive model. This logistic regression analysis revealed that 2 predictors - high pass vestibular roll gain (EAR UP) and low pass vestibular roll gain (EAR DOWN) - discriminate VN neurons from pmRF neurons with a sensitivity of 74% and specificity of 50%. pmRF neurons were more likely to have rectified high pass roll gain responses while VN neurons were more likely to have higher gain responses to low pass roll position signals. The present findings support the notion that neurons in vestibulospinal and reticulospinal pathways have complimentary functional roles in postural control. While neurons that integrate limb proprioceptive and vestibular signals in an additive manner likely govern corrective limb movements in response to postural perturbations through modulation of corrective reflexes, pmRF neurons may be more likely to be involved in modulating muscle activity during dynamic tasks (such as walking).

Disclosures: D.M. Miller: None. **C.D. Balaban:** None. **A.A. McCall:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.13/JJ28

Topic: D.08. Vestibular System

Support: FP7-624158

Title: Peripheral and central processing during vestibular unisensory integration in humans

Authors: *P. A. FORBES[1,2,3], G. P. SIEGMUND[4], A. KWAN[5], D. E. MITCHELL[6], A. C. SCHOUTEN[7], K. E. CULLEN[5], J.-S. BLOUIN[2]
[1]Dept. of Neurosci., Erasmus Med. Ctr., Rotterdam, Netherlands; [2]Univ. of British Columbia, Vancouver, BC, Canada; [3]Delft Univ. of Technol., Delft, Netherlands; [4]MEA Forensic Engineers & Scientists, Richmond, BC, Canada; [6]Dept. of Physiol., [5]McGill Univ., Montreal, QC, Canada; [7]Delft Univ. Tech., Delft, Netherlands

Abstract: Effective sensory integration arising from simultaneous biological events is essential to understanding and engaging with our surroundings. In the vestibular system, this integration typically results in subadditive responses, and can occur during multisensory (canal-otolith) and

unisensory (canal-canal) integration. Subadditive unisensory integration occurs due to a static-boosting nonlinearity in the input-output relationship of vestibular nuclei neurons, a process thought to expand their dynamic linear range. Understanding how vestibular unisensory integration modulates motor output in humans is critical for developing experimental, clinical and virtual/altered-reality applications that combine different forms of vestibular stimuli. In this study, we explored how electrical and motion vestibular stimuli are integrated to generate human neck muscle responses with the aim to determine at what neural stage integration occurs. We first applied electrical vestibular stimulation (EVS) as a stochastic signal (0-75 Hz) to seated subjects who were rotated about a vertical axis at 1 Hz with increasing peak amplitudes (0-150 °/s). Bilateral EMG of the sternocleidomastoid was collected to determine whether combined stimuli modified the electrically- and motion-evoked vestibulocollic reflex (VCR). We observed that the electrically-evoked VCR decreased across the stimulus bandwidth with increasing motion velocity. The timing of attenuation varied within the motion cycle and was similar in phase to the motion-evoked VCR: leading input velocity by ~40-60°. To further examine this relationship, we varied the frequency of motion from 0.4 to 4 Hz. Our results suggest that the phase of attenuation matched the dynamics of first and second order vestibular neurons, indicating that integration occurred prior to convergence on the motoneuron. To confirm that electrical and motion stimuli combine linearly at the afferent level, we then recorded macaque afferent responses during equivalent electrical and motion (1 Hz) stimuli used in humans. Afferent responses to the electrical stimulus were not altered by simultaneous whole-body motion. Instead, afferent activity could be predicted by the sum of each response, suggesting integration occurs at the vestibular nuclei. Finally, a model simulating the static-boosting nonlinearity of vestibular nuclei neurons replicated human neck muscle responses. Taken together, these results reveal that vestibular contributions to neck muscle activity evoked by electrical and motion stimuli undergo similar nonlinear integration within vestibular nuclei that occur during unisensory vestibular stimulation.

Disclosures: **P.A. Forbes:** None. **G.P. Siegmund:** None. **A. Kwan:** None. **D.E. Mitchell:** None. **A.C. Schouten:** None. **K.E. Cullen:** None. **J. Blouin:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.14/JJ29

Topic: D.08. Vestibular System

Support: NIH: R01DC009031 (HSC)

National Space Biomedical Research Institute through NASA NCC 9-58 (APM, JJB)

Fellowship from the Austria Marshall Plan Foundation (JS)

Title: Updated screening for vestibular and balance disorders

Authors: *H. S. COHEN[1], A. P. MULAVARA[2], B. T. PETERS[2], C. MILLER[2], J. STITZ[3], H. SANGI-HAGHPEYKAR[4], S. P. WILLIAMS[5], J. J. BLOOMBERG[6]
[1]Baylor Col. Med., Houston, TX; [2]KBRwyle, Houston, TX; [3]Univ. of Applied Sciences/ Upper Austria, Linz, Austria; [4]Obstetrics and Gynecology, [5]Med., Baylor Col. of Med., Houston, TX; [6]NASA/ Johnson Space Ctr., Houston, TX

Abstract: Our previous work studied one manner of testing tandem walking and the Romberg on foam with head still and head moving in pitch and showed moderate ROC values, sensitivity and specificity. Using larger sample sizes, (controls, n= 355; patients, n=140) we again tested Romberg on foam with eyes closed and feet together, with head still, head moving in yaw and in pitch at 0.3 Hz, controlled with an oscillating tone. We also tested subjects on tandem walking with eyes closed (controls, n = 291; patients, n=92). Logistic regression was performed for estimation of area under the curve (AUC), where cut offs for best combinations of sensitivity/specificity were determined. P <.05 was considered significant. Trial durations of Romberg pitch and yaw trials in controls were significantly shorter than head still trials but did not differ from each other, and trial durations decreased gradually and significantly with age. ROC values were moderate, depending on the exact cut of the age range, but were approximately 0.75. Tandem walking was tested with eyes closed, but instead of counting the number of correct consecutive steps the total number of correct steps out of ten was counted, on three consecutive trials. Subjects seemed to improve slightly on the second trial but fatigued on the third trial. Therefore, the first trial seems to be a good indicator of performance. ROC values were moderately high, better for younger subjects, < age 60, than subjects > age 60: ROC=0.8 younger subjects but ROC=0.72 older subjects. Kinematic data may provide additional insight but are not useful for screening.

Disclosures: H.S. Cohen: None. **A.P. Mulavara:** None. **B.T. Peters:** None. **C. Miller:** None. **J. Stitz:** None. **H. Sangi-Haghpeykar:** None. **S.P. Williams:** None. **J.J. Bloomberg:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.15/JJ30

Topic: D.08. Vestibular System

Support: Grant No. 81601639

Title: Reduction of motion sickness through targeting histamine N dash methyltransferase in the dorsal vagal complex

Authors: *H. L. XU, L. CHANG, Z. JIANG
Inst. of Nautical Med., Nantong Univ., Jiangsu, China

Abstract: Abbreviations: CNS, central nervous system; DVC, dorsal vagal complex; GFP, green fluorescent protein; HNMT, histamine N-methyltransferase; MS, motion sickness; SSS, saccharin sodium solution.

Abstract

To investigate the role of histamine N-methyltransferase (HNMT) activity in the development of motion sickness (MS) in the dorsal vagal complex (DVC) to inform the development of new drugs for MS, Beagle dogs and Sprague-Dawley rats were rotated to simulate MS. HNMT expression in the brain stem and DVC was measured. In addition, the effects of systemic application of tacrine, a HNMT inhibitor, on the development of MS were observed. Moreover, we microinjected a histamine receptor H1 inhibitor, promethazine, into the DVC to verify the involvement of DVC histaminergic neurotransmission in MS development. Finally, lentiviral vectors were microinjected into the DVC to determine the effects of altered HNMT expression on MS development. We found: 1) HNMT expression in the medulla oblongata of dogs and rats insusceptible to MS was higher than susceptible animals. 2) Tacrine dose-dependently promoted MS in both animals. 3) Blocking histaminergic neurotransmission in the DVC with promethazine inhibited MS. 4) Rotatory stimulus induced an elevation in HNMT expression and vestibular training elevated basal level of HNMT in the DVC during habituation to MS. 5) *In-vivo* transfection of a lentiviral vector packaged with HNMT gene increased HNMT expression in the DVC and reduced MS. 6) Microinjection of a lentiviral vector driving interference of HNMT gene expression *in vivo* significantly inhibited HNMT expression in the DVC and exacerbated MS. In conclusion, HNMT expression in the brain stem is inversely correlated with MS development. Increasing HNMT expression or stimulating its activity in the DVC could inhibit MS.

Disclosures: H.L. Xu: None. **L. Chang:** None. **Z. Jiang:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.16/KK1

Topic: D.08. Vestibular System

Title: Vestibular dysfunction impairs visuospatial working memory independently of other co-morbid depression, anxiety, fatigue and sleep disturbance

Authors: *S. SURENTHIRAN[1], L. J. SMITH[2], D. T. WILKINSON[2], R. BICKNELL[2], M. BODANI[2]

[1]Balance Centre, Neurosci. Unit, Medway Maritime Hosp., Gillingham, United Kingdom; [2]Sch. of Psychology, Univ. of Kent, Canterbury, United Kingdom

Abstract: Background Beyond the acute effects of vertigo and unsteadiness, disease or injury to the human vestibular system is commonly accompanied by subjective reports of memory loss and problems concentrating. The co-morbid presence of psychiatric illness, fatigue and difficulty sleeping, coupled with the lack of comprehensive, validated neuropsychological assessment, has left questions unanswered about the origin and nature of these underlying memory and attentional impairments. **Methods** 100 patients diagnosed with primary vestibular disorder (mostly vestibular migraine) at their initial neuro-otology appointment completed validated neuropsychological assessments of depression, anxiety, depersonalisation, fatigue, sleep, memory and attention. Vestibular pathology was characterised using a range of behavioural and physiological assessments. Statistical analyses first calculated the prevalence of cognitive and other comorbid impairments. A series of structural equation models then tested whether vestibular function exerted a direct influence on cognition, or influenced performance indirectly via psychiatric, fatigue/ sleep mechanisms. All models were adjusted for age-related effects on cognition. **Results** The majority of patients presented with a combination of anxiety, depression, sleep disturbance, fatigue, working memory impairments and problems sustaining attention. Most importantly, balance function, assessed via balance platform (a measure of unassisted posture), influenced visuospatial memory performance independently of any age, psychiatric or fatigue/ sleep-related effects. **Conclusion** The present findings identify new clusters of impairment in vestibular patients and highlight a direct effect of vestibular dysfunction on short-term visuospatial memory. We suggest that the most likely anatomical route is via the vestibulo-thalamo-cortical pathway which passes vestibular signals to several areas associated with working memory and visuospatial processing including the hippocampus, parietal cortex, frontal cortex and basal ganglia, From a clinical perspective, the results suggest that psychiatric treatments may do little to reduce co-morbid cognitive symptoms since they arise independently of psychiatric co-morbid factors and directly from the vestibular dysfunction.

Disclosures: S. Surenthiran: None. **L.J. Smith:** None. **D.T. Wilkinson:** None. **R. Bicknell:** None. **M. Bodani:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.17/KK2

Topic: D.08. Vestibular System

Support: NIH R01 DC004260

Title: Resolving the paradox of active versus passive self-motion sensation: A unified internal model theory

Authors: *J. LAURENS[1], ***J. LAURENS**[1], D. E. ANGELAKI[2]
[1]Neurosci., [2]Dept of Neurosci., Baylor Col. of Med., Houston, TX

Abstract: We present a theoretical framework that reconciles two segregated and seemingly conflicting lines of research on how the brain estimates self-motion during active and passive head movements. On one hand, brainstem and cerebellar neurons have been shown to implement an internal model of self-motion to accurately estimate rotations and translations from ambiguous and often inaccurate sensory vestibular signals during unpredicted, externally-generated ('passive') head and trunk movements. On the other hand, neurons in these same areas show reduced or no responses during self-generated ('active') head movements, a finding that suggests that the sensory consequences of motor commands are subtracted from sensory signals. Remarkably, the computational processes underlying sensory prediction during active motion and their relationship to internal model computations established during passive movements have never been modeled.
Here we show that the same internal model of self-motion described previously for passive vestibular stimuli should also process efference copies of motor commands in order to generate accurate sensory predictions. We provide support for this hypothesis by constructing a Kalman filter that incorporates motor commands into a previously-established Bayesian model of passive self-motion estimation. The Kalman filter computes sensory prediction dynamically based on internal estimates of head motion and motor commands, and converts sensory prediction errors into feedback signals that update the internal motion estimates. We simulate active and passive head rotations, translation, and combined head and trunk rotations, and find that the simulated feedback signals match neuronal responses recorded in the brainstem and cerebellum during active and passive rotation of the head and trunk, and translation of the head. We also predict neuronal responses to active and passive tilt, and long-duration active rotations and translation. We conclude that a single internal model of head motion, likely implemented by brainstem and cerebellar neuronal networks, can process motor commands and sensory afferent signals optimally, and that the Kalman model can provide valuable insights into the underlying neuronal computations as well as accurate quantitative simulations.

Disclosures: J. Laurens: None. **D.E. Angelaki:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.18/KK3

Topic: D.08. Vestibular System

Support: National Research Foundation of Korea (NRF), funded by the Ministry of Education, Science and Technology (2016R1A2B1012)

a faculty research grant of Yonsei University College of Medicine (6-2016-0092)

Title: Correlation of postural instability during virtual reality immersion to virtual sickness symptoms

Authors: *E. SON, K. ROH, J. KIM, J. KIM, S. HONG, S. KIM
Yonsei Univ. Col. of Med., Seoul, Korea, Republic of

Abstract: Virtual sickness refers to motion sickness-like symptoms elicited by immersion in virtual reality (VR) environment. VR technology can render 3D environment and is used in various industrial and medical applications and entertainment. However, a portion of users are deterred from VR by virtual sickness. Sensory conflict and postural instability theories are two main explanation given for virtual sickness. In this study, we aimed to investigate postural instability during VR experience and examine its correlation with subjective virtual sickness symptoms. Forty young subjects performed postural tasks adapted from modified clinical test of sensory integration test (standing on firm and foam surfaces) under four visual conditions (eyes open without VR, eyes closed, with VR goggles on, or while viewing a virtual reality scene). Postural instability was measured using center of gravity (COG) sway velocity, and subjective symptoms were measured using visual analog scale(VAS) and simulator sickness questionnaire(SSQ). The subjects showed a wide range of subjective symptoms related to virtual sickness measured using VAS and SSQ. Immersion in VR caused increased COG sway velocity compared to eyes open or closed conditions. COG sway velocity is increased sequentially from the eyes open, closed, goggle only and VR. COG sway velocity is also increased in foam surface conditions compared to firm surface. The effect of goggles was comparable to eyes closed conditions in both firm and foam surfaces ($P>0.5$). Postural instability was correlated with total and subscale (nausea, oculomotor, disorientation) scores of SSQ. Significant correlation with all subcales and total scores of SSQ was observed during VR only in Firm surface condition. In summary, even normal healthy subjects experienced variable levels of motion sickness during postural tasks in the VR immersion condition. Correlation of subjective symptoms with Increased postural sway during VR immersion supports the theory of postural instability contributing to virtual sickness.

Disclosures: E. Son: None. **K. Roh:** None. **J. Kim:** None. **J. Kim:** None. **S. Hong:** None. **S. Kim:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.19/KK4

Topic: D.08. Vestibular System

Title: The neural encoding of active self-motion by the primate cerebellum-evidence for an internal model that accounts for gravity

Authors: *I. MACKROUS[1], J. CARRIOT[2], K. E. CULLEN[3]
[1]Physiol., McGill, Montreal, QC, Canada; [2]Physiol., [3]McGill Univ., Montreal, QC, Canada

Abstract: During daily activities, our sensory system is activated by self-generated and external events. We have shown that cerebellar output neurons robustly encode passively applied motion in the horizontal plane, while they are attenuated during comparable self-generated head motion. However, the complex trajectory of natural head motion poses a particular challenge for the vestibular system because of the presence of the gravity. This is because the sensory periphery does not provide an explicit representation of gravity. Here we asked whether an estimate of gravity is included in the internal model used to compute the difference between self-generated and external self-motion by studying the neuronal responses of cerebellar output neurons (rostral Fastigial nuclei, rFN) during active motion. Based on their responses to passive motion, neurons could be divided in three groups: 1) GIA selective cells that respond similarly to tilt and translation, 2) tilt-selective cells that showed greater responses to tilt, and 3) translation-selective cells that showed greater responses to translation. We then recorded the responses of each individual neuron during conditions in which monkeys actively changed their head orientation (tilts) or made active head translations. Notably, the responses of the GIA and the tilt-selective cells were attenuated during active head tilts (80%). Furthermore, translation-selective cells showed a ~80% attenuation in their neuronal response during active compared to passive translation, and were also insensitive to active tilts. Thus, changes in head orientation relative to gravity were not encoded at the rFN level by any cell type during active motion. This finding implies that the generation of the required cancellation signal is based on an internal model of the expected sensory consequences that accounts for gravity. To further test whether the cancellation signal was specific to the active motion, or instead a more general gating mechanism, we also recorded neural response during passive translation while the monkeys made simultaneous voluntary head tilt motion. When submitted to concomitant stimuli, neuron's response to the active tilt stimulus was attenuated while cells' responses provided a precise estimate of the passive translation motion. Thus, taken together our findings demonstrate the output of a sophisticated computation, in which the influence of gravity is taken into account. Specifically, we propose a new model, which includes an estimate of the influence of gravity during voluntary motion, to explain how the brain differentiates between self-generated and passively applied head movements.

Disclosures: I. Mackrous: None. **J. Carriot:** None. **K.E. Cullen:** None.

Poster

404. Vestibular System: Central Processing

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 404.20/KK5

Topic: D.08. Vestibular System

Title: Inter-hemispheric control of vestibular thresholds

Authors: *N. BEDNARCZUK, M. CASANOVAS-ORTEGA, A.-S. FLURI, A. M. BRONSTEIN, Q. ARSHAD
Dept. of Neuro-otology, Div. of Brain Sci., Imperial Col. London, London, United Kingdom

Abstract: Neural mechanisms underpinning vestibular cortical control are subject to inter-hemispheric interactions, which in turn can modulate both low and higher order vestibular functions. However, whether inter-hemispheric interactions can influence vestibular thresholds remains unknown. Hence, we investigated whether individual differences in hemispheric dominance, as assessed using a validated, objective biomarker, could predict changes in vestibulo-ocular (VO) and vestibulo-perceptual (VP) thresholds following alterations in cortical excitability using trans-cranial Direct Current Stimulation (tDCS). Twenty right-handed normal subjects were recruited, with ten subjects receiving cathodal stimulation (experimental condition), whilst the remaining ten received anodal stimulation (control condition). Stimulation was applied with a current of 1.5mA for 15min over the left posterior parietal cortex (P3, 10-20 international EEG system), with the reference electrode placed on the ipsilateral shoulder. Hemispheric dominance was quantified by assessing the degree of vestibular-nystagmus suppression in response to cold-water (30 ^{o}C) caloric irrigations in both ears after tDCS. To measure vestibular thresholds, participants were rotated in a vibration-free, motorised chair with an initial velocity of 0.3^{o}/s and an incremental acceleration of 0.3 ^{o}/s^{2}. Rotations occurred in complete darkness with amplified white noise to mask auditory cues, and were stopped as soon as subjects pressed the direction-specific button, indicating motion perception. Simultaneously, eye movement recordings were obtained using electro-oculography. Analysis of this data provided VO thresholds, which reflect the time taken from onset of chair rotation to the first nystagmic beat, and VP thresholds, which represent the time taken from rotation onset to the participant's button-press. After cathodal stimulation, the average VO thresholds became symmetrically raised, whilst average VP thresholds remained largely unchanged. Importantly, we observed a significant, positive correlation between individuals' hemispheric dominance and the degree of asymmetry present between right and left VO thresholds. Hemispheric dominance was also predictive of an individual's degree of change in VP thresholds following cathodal tDCS, but only for rightward and not leftward rotations. We provide a novel demonstration of the importance of inter-hemispheric interactions in modulating vestibular thresholds following alterations in cortical excitability using tDCS.

Disclosures: **N. Bednarczuk:** None. **M. Casanovas-Ortega:** None. **A. Fluri:** None. **A.M. Bronstein:** None. **Q. Arshad:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.01/KK6

Topic: E.04. Voluntary Movements

Support: Office of Naval Research award N000141310597

Lockheed Martin Corporation

Title: High-level motor planning assessment during performance of complex actions in humans and humanoid robots: A computational approach

Authors: ***T. C. HAUGE**[1], G. KATZ[2], D.-W. HUANG[2], G. DAVIS[2], J. A. REGGIA[2], R. J. GENTILI[3]
[1]Univ. of Maryland, Col. Park, College Park, MD; [2]Computer Sci., [3]Kinesiology, Univ. of Maryland, College Park, MD

Abstract: Most past work on cognitive-motor processes underlying upper-extremity motor control and learning has examined the sensorimotor level while considering relatively simple movements. However, complex cognitive-motor planning is needed to generate actions of daily living that are composed of different sub-actions. Therefore, in addition to kinematic analyses that can assess sensorimotor processes, there is a critical need to develop formal methods for representing and analyzing complex action sequences to better understand high-level motor planning. Here we describe a general computational method for comparing complex tasks performed by humans and humanoid robots. Our approach is based on sequence comparison methods that are employed in linguistics and genomics, but now the atomic elements (nodes) represent the individual actions forming complex action sequences. Such an approach allows one to examine qualitatively (graphical representations) as well as quantitatively (clusters of similar trials; similarity metrics) the structure of complex actions. For assessment purposes, the proposed approach was tested on data that were collected during a complex imitation learning task, in which human participants as well as a humanoid robot learned a disk-drive dock maintenance task, based on a demonstration recorded in a virtual human-operated environment. Once learning was complete, the humanoid robot was able to generalize the task to novel initial states by inferring the demonstrator's intentions. Then, human participants had to imitate the same task demonstrated in a video with minimal instruction. The results revealed that for most trials, human participants were able to successfully imitate the demonstrated goal of the task. However specific differences between the demonstration, humans, and humanoid robot actions were

observed suggesting that different strategies were employed for achieving the same goal. Specifically, our computational method was able to detect: i) identical and modified sequences; ii) unique sequences performed by humans, iii) the distribution of trials among sequences and iv) levels of similarity between actions performed by the human demonstrator, the human participants, and the humanoid robot. We conclude that this is a promising tool for analyzing high-level motor planning in humans during cognitive-motor control and learning of complex actions as well as for informing human-robot interaction studies.

Disclosures: **T.C. Hauge:** None. **G. Katz:** None. **D. Huang:** None. **G. Davis:** None. **J.A. Reggia:** None. **R.J. Gentili:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.02/KK7

Topic: E.04. Voluntary Movements

Support: JSPS KAKENHI 26120004

JSPS KAKENHI 25290001

Title: Postural control that precedes to the forelimb reaching in the cat

Authors: *K. TAKAKUSAKI[1], M. TAKAHASHI[1], S. MIYAGISHI[1], T. DREW[2]
[1]Asahikawa Med. Univ., Asahikawa/Hokkaido, Japan; [2]Dept. of Neurosci., Montreal Univ., Montreal, QC, Canada

Abstract: Appropriate postural control precedes the onset of the purposeful action so that the subject achieves goal-directed movements. Such a feed-forward postural control is often called as the anticipatory postural adjustment (APA). Although evidence was accumulated on the mechanisms of cortical control of APA, there have been few studies that have specifically asked how APA ensure goal-directed movements. To understand this issue, postural control associating with forelimb reaching was examined in four unrestrained cats weighing from 2.2 to 3.3 kg. Cats were trained to stand by their four-limbs toward the panel which contained several holes (targets) at various positions. The cats were then asked to reach either of their forelimbs to one of the targets and catch food pellets inside of the holes. The cat movements were monitored by video-tracking systems, and ground reaction forces impinging on each limb was measured by force transducers so that changes in center of vertical pressure (CVP) against left-right and anterior-posterior coordinates were examined. Forelimb reaching movement was divided into three phases. The first phase was the onset of APA which was typically identified by a loading of the forelimb to be lifted. With this, CVP moved toward ipsilateral to the forelimb side. This was

immediately followed by the second phase which was characterized by the period of unloading of the forelimb until the forelimb was apart from the surface. During this phase, the cat poked up one's head together with leaning trunk toward contralateral side of the forelimb. During such a postural alteration, the CVP was rapidly shifted contralateral side of the forelimb. It takes approximately 0.25 - 0.3 seconds for the first and second phases. The third phase was a period for reaching movement until the forelimb touch the target after the plantar was lift from the surface. This phase took approximately 0.2 - 0.3 seconds. Although CVP was altered in conjunction with forelimb reaching, CVP position at reaching the target was mostly equal to the CVP position at the end of the second phase. When position of the target was changed from the original position to either the left or right, CVP positions of the second and third phases was altered in relation to the target position. These findings suggest that APA provides the final stage of posture that can be optimized to ensures the achievement of voluntary forelimb reaching. Therefore, this process requires motor programs that depend on cognitive spatial information of one's body and the target. It can be postulated that the APA is mediated by descending motor systems other than the lateral corticospinal tract such as the cortico-reticulospinal pathways.

Disclosures: K. Takakusaki: None. **M. Takahashi:** None. **S. Miyagishi:** None. **T. Drew:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.03/KK8

Topic: E.04. Voluntary Movements

Support: NIH Grant T32GM081740

USC Office of Research ASPIRE I Award

Title: Effect of practice on the control of reach extent

Authors: *J. C. STEWART, A. HETHERINGTON, D. BRUEMMER, T. ICHIYANAGI, J. ROCKTASHEL, M. O'DONNELL, C. SIMMONS, T. M. HERTER
Univ. of South Carolina, Columbia, SC

Abstract: The systematic scaling of reach kinematics to targets that vary in distance has been well-described. To control reach extent, individuals generally utilize a control pattern that includes both anticipatory planning and feedback based adjustments. However, the stability of this control pattern over days of practice is not known. The purpose of this study was to determine the effect of three days of practice on the control of reach extent for reaches to targets presented in 3-dimenionsal space. Twenty-four young, right-hand dominant adults (mean age

23.4 ± 2.7) performed reach movements to 3-dimensional targets that varied in distance (7, 14, 21 cm) and direction (ipsilateral, contralateral workspace) over 3 days of practice; 12 individuals reached with the dominant, right arm and 12 individuals reached with the nondominant, left arm. Position data was captured with a sensor on the index finger and used to extract kinematic variables of reach performance (movement time, endpoint error, velocity, acceleration) and the reach control pattern (planning-variance in reach distance explained by peak acceleration magnitude; feedback based adjustments-variance in reach distance explained by acceleration duration). Reach performance improved over practice (increased peak velocity, decreased movement time, decreased error) in both Right and Left arm groups ($p<0.05$) across target directions. Both groups showed a control pattern that include both planning and feedback adjustments; the overall control pattern was similar to ipsilateral and contralateral targets. Over days of practice, the Left arm group showed a significant increase in the use of planning and adjustments in both directions ($p<0.05$), however, the magnitude of changes was relatively small (increase in percent variance explained ranged from 3 to 7%). The Right arm group did not show any significant changes in the reach control pattern in either direction with practice. Consistent with previous work, reach performance improved with practice for measures of both reach speed and reach accuracy, even for this relatively simple task. The change in the control pattern seen with the left arm but not the right arm may be related to dominance. The right arm was the dominant arm in all participants; this arm may have a more stable control pattern than the nondominant, left arm for the control of reach extent due to overall greater frequency of use in everyday activities. Future work will investigate the effect of practice on the control of reach extent in individuals with reach control deficits.

Disclosures: J.C. Stewart: None. **A. Hetherington:** None. **D. Bruemmer:** None. **T. Ichiyanagi:** None. **J. Rocktashel:** None. **M. O'Donnell:** None. **C. Simmons:** None. **T.M. Herter:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.04/KK9

Topic: E.04. Voluntary Movements

Title: Visual feedback may establish an implied context for reaching behavior

Authors: *S. G. PENNY, N. VAIDYANATHAN, M. BERNIKER
Mechanical and Industrial Engin., Univ. of Illinois At Chicago, Chicago, IL

Abstract: It is known that reaching movements can be influenced by altering visual feedback, but could visual feedback itself be biasing how reaches are controlled? Efficient reaches (i.e. those that minimize work or energy) should move the hand along curved paths, but studies

consistently find straight reaches. Does the visual feedback of standard reaching tasks implicitly cue subjects to perform a "visual capture" reach, moving the visual cue along a straight path to the target and disregarding any inherent preferences for efficiency? We hypothesized that if this were true, even a brief display of visual cues would bias how subjects reach. To test this, we had subjects participate in one of two reaching experiments: visual feedback is either introduced or removed halfway through testing. In experiment 1, continuous visual feedback of the hand and target location were provided during reaches for the first half of the trials, and then both hand location and targets were extinguished for the remainder of the trials. In experiment 2, feedback of neither the hand nor the targets were provided for the first half of the experiment, but were then revealed for the second half. To aid subjects during the extinguished visual feedback portion, the target number and an "error bar" (indicating the unsigned distance to the target) were displayed. In experiment 1, we found that subjects made straight movements while the visual feedback was provided, and continued to make straight reaches after visual feedback was removed. In experiment 2, subjects made curved movements while no feedback was provided, but immediately switched to straight movements once presented with the feedback. Our findings suggest that visual feedback may influence the planning of reaches, resulting in straight movements even after feedback is extinguished.

Disclosures: S.G. Penny: None. **N. Vaidyanathan:** None. **M. Berniker:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.05/KK10

Topic: E.04. Voluntary Movements

Title: Local generalization curves after force field adaptation

Authors: *A. REZAZADEH[1], M. BERNIKER[2]
[1]Dept. of Mechanical & Industrial Engin., [2]Mechanical and Industrial Engin., Univ. of Illinois At Chicago, Chicago, IL

Abstract: To understand how we learn, we often examine the ability to generalize. Thus, to study motor learning many studies probe the ability to make unpracticed reaches after adapting to a force field. This is often executed using reaches that are different (i.e. far away) from those practiced. Recent findings suggest that the ability to generalize is more limited and local than previously assumed; however, this evidence does not account for potential changes in limb inertia across reach directions. To address this, we designed a force field study to examine the extent of generalization while controlling for limb impedance. Subjects practiced reaches in a curl field to one of eight targets and were then tested in all directions using no-vision error-clamp trials. We computed generalization curves for each direction by pooling the data across all eight

reaches in order to correct for changes in limb impedance. These results provide a thorough evaluation of the localized ability to compensate for a force field and speak directly to how new motor behaviors are represented.

Disclosures: A. Rezazadeh: None. **M. Berniker:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.06/KK11

Topic: E.04. Voluntary Movements

Title: Are straight reaches the result of visual feedback?

Authors: *N. VAIDYANATHAN[1], S. PENNY[2], M. BERNIKER[1]
[1]Mechanical and Industrial Engin., Univ. of Illinois At Chicago, Chicago, IL; [2]Univ. of Illinois at Chicago, Chicago, IL

Abstract: While biologically relevant goals such as minimizing forces or joint torques dictate curved hand trajectory, reaching studies consistently find straight point-to-point hand paths. It is well documented that visual feedback affects how subjects reach, but could this influence explain the lack of curvature experimentally found? To address this question we reproduced an influential study (Morasso 1981) that argued point-to-point reaches are planned to move the hand's coordinates along straight paths, while moving the limb joints along indirect paths to their target. In the first protocol subjects were provided feedback of their hand location as well as targets. In a second protocol, feedback of the hand's location was replaced with a circle centered on the target, whose radius was equal to the subject's distance to the target; therefore, subjects were shown their distance to the target, but not their exact hand location. In a third protocol, feedback of both the hand and the target location was extinguished, and subjects were provided with the target number, and an error bar, indicating the unsigned distance to the target; here, subjects had to rely on the remembered target locations. As prior studies have shown, when the standard visual feedback of hand and target was provided, subjects moved their hand along relatively straight paths. But as visual feedback was attenuated subjects moved their hands along progressively curved paths. To help explain these changes and also to comprehend the strategies involved in reaching movements, a secondary analysis of the subject's limb orientation - involving the path of elbow and shoulder angles was performed. We suggest that as visual feedback is attenuated, and reaches rely less on the visual cues provided, the planning of reaches may prioritize limb dynamics over hand coordinates. Therefore, this experimental paradigm may unmask previously unexamined features of the control and planning of movement.

Disclosures: N. Vaidyanathan: None. **S. Penny:** None. **M. Berniker:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.07/KK12

Topic: E.04. Voluntary Movements

Support: NIH Grant # R01-AG049735

Title: Modeling a shuffleboard machine that regulates motor noise like a human

Authors: *J. P. CUSUMANO[1], C. J. SMITH[2], J. B. DINGWELL[3]
[1]Dept. of Engin. Sci. & Mechanics, [2]Engin. Sci. & Mechanics, Penn State Univ., University Park, PA; [3]Kinesiology, Univ. of Texas At Austin, Austin, TX

Abstract: In recent work with human subjects playing a virtual shuffleboard game, we showed that the analysis of trial-to-trial fluctuations near the task's goal equivalent manifold (GEM) sheds light on how humans exploit motor redundancy to achieve repeatable, accurate performance while coping with intrinsic physiological noise (John et al., PLoS Comput. Biol. 2016). Our model-based, dynamical approach yields a coordinate-independent characterization of the combined geometrical and temporal structure of motor variability in repeated trials, showing how goal-level task error arses from the interplay of motor noise, passive sensitivity properties of the GEM, and inter-trial error correction. However, our variability analysis uses models built from experimental end-effector data (for shuffleboard, the position and velocity of the puck at release), so it is difficult to directly relate the resulting abstract measures of performance (e.g., geometric stability, correlation) to the underlying neuro-biomechanical system generating it. To bridge this gap between experimental methods and first-principles, we formulated a physics-based model of a two-link shuffleboard machine driven by a motor with state-dependent torque, with pseudo-random noise in the motor output representing human physiological noise. We show how the shuffleboard GEM and its sensitivity properties transform between the space of end-effector variables and the space of internal torque parameters. Multi-trial simulations show that inter-trial error regulation that only aims to keep the system near a generic operating point with mean-zero goal-level error does not result in a variability structure like that observed in human shuffleboard players, whereas “GEM aware” error regulating controllers do. In particular, the dynamical anisotropy observed in human subjects experiments, in which inter-trial error regulation acts much more strongly in directions transverse to the task manifold than along it, is recovered in the model with GEM aware controllers. Furthermore, we show that the fluctuation analysis previously applied to human subjects, which uses only directly observable end-effector data, accurately characterizes the system dynamics as described in the space of its intrinsic motor variables.

Disclosures: J.P. Cusumano: None. **C.J. Smith:** None. **J.B. Dingwell:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.08/KK13

Topic: E.04. Voluntary Movements

Support: NIH NS092079

Title: Task dependent modulation of implicit visuomotor adaptation

Authors: *D. E. PARVIN[1], J. R. MOREHEAD[2], A. R. STOVER[1], R. B. IVRY[1]
[1]UC Berkeley, Berkeley, CA; [2]Sch. of Engin. and Applied Sci., Harvard Univ., Cambridge, MA

Abstract: Implicit sensorimotor adaptation is thought to be driven by discrepancies between 'predicted' and 'actual' feedback, or sensory prediction errors (SPEs). It has been suggested that this process is automatic and obligatory, even when detrimental to task success (Srimal *et al.*, 2008; Mazzoni & Krakauer, 2006). One explanation for this apparent immunity to task success is that sensory predictions are generated relative to the efferent motor command, irrespective of the context in which this command was generated. To explore whether implicit adaptation is truly independent from the task conditions, we developed a task that allowed us to manipulate the task context without altering the actual movements or the ostensible sensory prediction errors introduced by altered feedback. Specifically, we used multiple redundant feedback cursors to show a participant's hand position in a center-out planar reaching task (Kasuga, Hirashima & Nozaki, 2013). The redundancy of the feedback allowed separate groups of participants to perform reaching movements with the same visuomotor mapping but under different task instructions specifying the task-relevant cursor. After the learning block, we probed implicit adaptation and generalization by having the participants moved their unseen hand directly to the target location without visual feedback. In all baseline conditions, each of the three cursors had a constant rotation of -45º, 0º and 45º, and participants were instructed to hit the target with the 0º cursor. For the learning blocks, we created conditions based on the overall rotation of the three cursors and the rotation of the task-relevant cursor, the one they were instructed to hit the target with. In the first group, we kept the overall cursor rotation as in baseline, but changed the task-relevant cursor to the 45º one. In the other two groups, we rotated the overall cursor feedback by 45º, resulting in cursors with rotations of 0º, 45º and 90º. In the second group and third group, the task-relevant cursor was at 0º and 45º respectively. The aftereffects depended on the rotation of the task relevant cursor, rather than the overall rotation of the three cursors. These results indicate that the motor system intelligently modulates the influence of putative error signals for learning in the context of redundant feedback. This is consistent with either a modulation via visual attention, or of a dedicated selection system for visuomotor binding (Reichenbach et al 2014) which links the hand representation in visual and motor systems.

Disclosures: **D.E. Parvin:** None. **J.R. Morehead:** None. **A.R. Stover:** None. **R.B. Ivry:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.09/KK14

Topic: E.04. Voluntary Movements

Support: CIHR

NSERC

Title: Gaze patterns provide a read out of strategy use in visuomotor adaptation

Authors: *A. J. DE BROUWER, M. ALBAGHDADI, J. R. FLANAGAN, J. P. GALLIVAN
Queen's Univ., Kingston, ON, Canada

Abstract: Sensorimotor adaptation is thought to reflect the resultant combination of two separate, but interacting processes: the implicit adaptation of an internal model, and the use of an explicit, cognitive strategy. Current evidence suggests that explicit strategies underlie faster adaptation (i.e., savings) when re-exposed to the same perturbation a second time. Interestingly, previous research indicates a link between gaze behavior and the explicit process (Rand & Rentsch, 2015). Here, we systematically examined gaze behavior over the time course of adaptation and re-adaptation to a visuomotor rotation, and directly tested the idea that gaze fixations provide a read out of the state of explicit adaptation during learning. Two groups of participants performed two sessions (separated by 24 h) of fast center-out reaching movements while adapting to an instantaneous 45° rotation of the visual feedback of the hand cursor. Participants in the intermittent report group were asked to verbally state the direction they would move their hand in before starting the movement (see Taylor et al., 2014) on one quarter of the trials. Participants in the non-report group were not required to report their aiming direction. We found that gaze fixation angles preceding the reach movement were bimodally distributed for 18 out of 21 participants in the intermittent report group and for 13 out of 21 participants in the non-report group. One peak was located at the visual target location, and the second peak was closely aligned to the explicit component, as measured in verbal reporting trials in the intermittent report group. Notably, in the non-report group, we observed faster adaptation and greater savings in participants that naturally fixated an internal aimpoint, indicating the implementation of an explicit strategy. Together, these results suggest that gaze fixations, prior to reaching, can be used to probe cognitive strategies during sensorimotor learning and utilized as a substitute for verbal reporting measures, which may influence adaptation.

Disclosures: **A.J. De Brouwer:** None. **M. Albaghdadi:** None. **J.R. Flanagan:** None. **J.P. Gallivan:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.10/KK15

Topic: E.04. Voluntary Movements

Support: CIHR project grant MOP-142220

Title: PMd reach-related activity expresses a response component related to the strength of evidence used to choose a target before, during and after the movement

Authors: *C. MONTANEDE[1], J. F. KALASKA[2]
[1]Neurosciences, Univ. of Montreal, Montreal, QC, Canada; [2]Neurosciences, Univ. De Montréal, Montréal, QC, Canada

Abstract: Monkeys pointed to either a Blue or Yellow target in opposite directions according to their estimate of whether a multi-colored checkerboard-like Decision Cue (DC) had more B or Y squares (Choose-and-Go (CG) task; Coallier and Kalaska 2014). The DCs presented in different trials had different total numbers and differences in numbers (color bias) of B and Y squares. Neurons in dorsal premotor cortex (PMd) often showed a graded rate of increase in activity before reach movement onset as a function of the strength of the color bias in the DCs (Coallier et al 2015). We also noted that some PMd neurons continued to show color bias-dependent differences in activity during the movements and while holding the arm over the targets. Here, we confirm that latter property in a new monkey. We recorded activity of 59 PMd neurons near the genu of the arcuate sulcus in the CG task. We tested the responses for the effects of chosen target direction (PD, oPD), and the color bias (4-100 squares) and predominant color (B, Y) of DCs (repeated-measure 3-way ANOVA, $p<0.01$). Most neurons showed a significant main effect of direction (Reaction Time epoch: 43/59; Movement Time epoch: 50/59; Target Hold epoch: 44/59). Many also showed a significant main effect of color bias (systematic increase or decrease of discharge rate in both directions as a function of bias; RT: 13/59; MT: 24/59; TH: 31/59). A partially overlapping set of neurons showed a significant interaction between direction and bias (significant differences in activity as a function of bias that differed for the 2 reach directions; RT: 7/59; MT: 10/59; TH: 9/59). Far fewer neurons showed a significant main effect of predominant color (0/59; 7/59; 4/59) or direction-color interaction (3/59; 2/59; 2/59). Similar trends were seen for the same neurons in CG task variants in which a delay was imposed between the presentation of the DCs and a later GO signal, or in which the monkey saw the DC first for 1-2s before being shown the 2 targets. Response differences were much less prominent when trials were sorted according to reach reaction time rather than color bias. The effect of

color bias was much weaker in a set of neurons recorded in primary motor cortex close to the central sulcus (e.g., main effect – RT: 7/26; MT: 7/26; TH: 3/26). These findings suggest that PMd reach-related activity expresses a response component that covaries with the strength of evidence (color bias independent of predominant color) on which the monkey made its target choice for the entire duration of the trial, not just before movement onset. This component may reflect various factors such as the level of confidence of the monkey in its target choice, reward expectations or error detection.

Disclosures: C. Montanede: None. **J.F. Kalaska:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.11/KK16

Topic: E.04. Voluntary Movements

Support: 4R01HD073147-05

JSPS Overseas Research Fellowships

Title: Reinforcement learning leads to increase in exploration variability

Authors: *S. UEHARA[1,4], F. MAWASE[1], A. S. THERRIEN[2,5], K. M. CHERRY-ALLEN[1], A. J. BASTIAN[2,5], P. CELNIK[1,2,3]
[1]Dept. of Physical Med. and Rehabil., [2]Dept. of Neurosci., [3]Dept. of Neurol., Johns Hopkins Univ., Baltimore, MD; [4]Japan Society for the Promotion of Sci., Tokyo, Japan; [5]Ctr. for Movement Studies, Kennedy Krieger Inst., Baltimore, MD

Abstract: Exploration is thought to serve a critical role in motor learning, allowing us to find the optimal motor solution to perform a new task. In particular, exploration seems to be critical in reinforcement learning processes because it allows the motor system to determine which movements lead to the most reward. Indeed, previous studies have shown that exploration variability influences performance in reinforcement-based learning tasks (Wu et al. 2014; Therrien et al. 2015). However, it remains unclear whether the process of reinforcement learning can also lead to changes in exploration. Here we show that exploration variability is integral to reinforcement learning, but that reinforcement learning also shapes exploration.
In Experiment 1, we evaluated the magnitude of exploration variability while participants learned a new goal-directed finger pointing movement using only binary feedback about their movement success (vision of their movements was occluded). As a proxy for exploration variability, we measured trial-to-trial changes in pointing angle as a function of whether the preceding trial resulted in success or failure. The magnitude of movement angle change after

each failure trial was used as the measure of exploration. We found that exploration gradually increased and persisted at high levels even after the pointing movements matched the predetermined target angle (i.e. the optimal motor solution was found). Participants who were unable to reach the target angle showed smaller exploration variability throughout the training. In Experiment 2, we investigated whether the increased exploration benefited performance in a subsequent exposure to the same task. A new group of participants performed the same motor task twice, with a washout phase in between the two learning phases. We found that participants showed faster learning in the 2nd phase, which was accompanied by greater exploration from the beginning of the 2nd phase. To control for potential directional biases in pointing angle after the 1st phase, we conducted a third experiment that was identical to Experiment 2, but in the 2nd phase participants learned pointing movements in the opposite direction to movements made in the 1st phase. We found that, similar to Experiment 2, learning of the task in the 2nd phase was faster than that in the 1st phase. This faster learning was also proportional to the magnitude of motor exploration.
These findings indicate that the motor system may flexibly regulate exploration variability during reinforcement learning, which facilitates learning of similar tasks. This indicates a bidirectional influence between exploration and reinforcement learning.

Disclosures: S. Uehara: None. **F. Mawase:** None. **A.S. Therrien:** None. **K.M. Cherry-Allen:** None. **A.J. Bastian:** None. **P. Celnik:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.12/KK17

Topic: E.04. Voluntary Movements

Support: NWO/STW Grant Symbionics 13524

Title: Are physically interacting partners mutually optimal?

Authors: *N. BECKERS, A. KEEMINK, E. VAN ASSELDONK, H. VAN DER KOOIJ
Univ. of Twente, Enschede, Netherlands

Abstract: Physically interacting partners have shown to improve motor performance, coordinate actions or select Nash equilibria in non-cooperative motor games. However, it remains unclear how humans change and shape their control policies during two-person physical interaction in cooperative movement tasks. Diedrichsen showed that feedback control changed optimally with task goals in bimanual reaching; a perturbation applied to only one hand was compensated optimally by both hands. In bimanual motor control the central nervous system has direct knowledge of the dynamics and task goals of both hands, allowing an optimal *centralized* control

strategy. However, physically interacting partners can only indirectly infer the dynamics of their partners through sensorimotor interaction to construct a control policy. In addition, partner dynamics are not time-invariant. If one partner changes control policy, this changes the dynamics the other partner interacts with. Since the control policy that is optimal for one interaction dynamics is unlikely to remain optimal for another, changed, interaction dynamics, the other partner will most likely reoptimize his/her control policy. Importantly, each partner only optimizes for their *own* cost function and inferred (closed-loop) partner dynamics. If this process occurs in physically interacting partners, partners will ultimately become *mutually optimal* with respect to each other's *separate* task goals, dynamics and control policies. However, if both partners manage to estimate the dynamics, state and cost function of their partners the mutually optimal controllers could select a control policy approaching a centralized controller, as in the bimanual case. Our objective is to test whether two physically interacting partners indeed change control policies such that they are mutually optimal controllers. Using a dual-interface robotic setup we couple two partners with a virtual spring. Both partners simultaneously perform reaching movements to the same target. A force field is then introduced for one of the partners to evoke reoptimization of control policies. By matching the measured force responses during catch trials to computational models with either two mutually-optimal controllers or an optimal centralized controller, we intend to test whether interacting partners select mutually optimal strategies, a centralized strategy or exploit a different strategy altogether. This work is useful in unraveling the mechanisms of physical interaction and to develop optimal human-robot interaction paradigms.

Disclosures: **N. Beckers:** None. **A. Keemink:** None. **E. van Asseldonk:** None. **H. van der Kooij:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.13/KK18

Topic: E.04. Voluntary Movements

Support: NIH Grant T32 HD007430

NIH Grant DP2 1DP2 MH10016-01

McKnight Grant MF CU13-0021

Simons foundation

Klingenstein-Simons Fellowship

Seattle Scholars Program

Kavli Foundation

Title: Preparatory activity in motor cortex and supplementary motor area does not reflect the instantaneous probability of choosing to move

Authors: *A. J. ZIMNIK, A. H. LARA, M. M. CHURCHLAND
Dept. of Neurosci., Columbia Univ., New York City, NY

Abstract: When deciding which movement to execute, activity in monkey motor and premotor areas can wax, wane, or vacillate in ways that reflect the instantaneous likelihood of making a particular choice. But does such activity relate directly to the decision process, or result from a strategy to prepare movement as soon as a choice becomes likely? To dissociate these possibilities, we employed a task where the decision of when to reach evolved over the course of seconds, guided by internal estimations and external cues. In contrast, the physical location of the reach target was certain upon stimulus onset. Neural activity reflecting the evolving likelihood of executing a choice should wax and wane with the measured probability of choice execution. Conversely, activity reflecting motor preparation should change rapidly upon stimulus onset but remain relatively constant with time. To achieve the desired dissociation, only one target was available on a given trial, but the decision of when to move had to take into account two factors: a monotonically increasing reward and a sensory cue conveying the fluctuating probability that the target might become unavailable. The monkey's probability of choosing to move appropriately reflected these two factors and could be predicted (R^2 = 84%) via an economic model. We recorded from motor cortex (primary and premotor cortex, 152 neurons) and from supplementary motor area (SMA, 242 neurons) and concentrated on activity during the decision period. We analyzed the top 2 principal components, which accounted for ~25% of the variance and were selective for target location. We expected these components to reflect the time-varying probability of choosing to move, such that activity selective for a particular direction would be stronger when the probability of moving in that direction was higher. Yet this was not the case. Selective activity emerged immediately upon target onset and remained strong regardless of whether the probability of moving was low or high. This observation is consistent with activity reflecting motor preparation; selectivity is fully present once the reach direction is known. We used regularized regression to ask whether there exist other components that correlate with the probability of choosing to move. Such components were present but captured little variance (4.9% and 3.8% in SMA and motor cortex). Our results are consistent with two possible interpretations of decision-period activity in motor and premotor areas. First, such activity may primarily reflect movement preparation, and lie downstream of the decision process. Second, such activity may reflect the decision of what movement to make but not when to make it.

Disclosures: **A.J. Zimnik:** None. **A.H. Lara:** None. **M.M. Churchland:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.14/KK19

Topic: E.04. Voluntary Movements

Support: CIHR Grant MOP-142220

Title: Relative sensitivity of subjects' performance to total, net and relative evidence in three color-quantity estimation tasks

Authors: *S. DUROCHER, J. MILOSZ, O. IERFINO, J. KALASKA
Neurosciences, Univ. De Montreal, Montreal, QC, Canada

Abstract: Many studies of two-alternative perceptual- and action-related decisions based on noisy signals (“evidence”) suggest that subjects optimize the decision process by sequential sampling of evidence to alter the value of a decision variable until it exceeds a decision threshold. One large class of models assumes that the decision variables are altered by the difference in momentary evidence (“net evidence”; NE) for each choice whereas another class assumes that the variables are modulated only by the absolute evidence for their respective choice (“total evidence”; TE). NE models have been extensively supported by studies in which subjects identify the coherent motion direction of a subset of dots in random-dot motion (RDM) stimuli. However, in nearly all cases, the net coherent motion was in only 1 of the 2 directions so that NE and TE is essentially identical. In contrast, two recent studies in which subjects identified the brighter of two visual stimuli over a wide range of different absolute and relative brightness showed a significant influence of the absolute brightness (i.e., TE) on subjects’ performance (Liston & Stone 2013; Teodorescu et al 2015). We tested to what degree subjects were sensitive to NE vs TE in 3 tasks in which they estimated whether there were more Blue (B) or Yellow (Y) squares in a multi-colored checkerboard (Decision Cue; DC) (Coallier & Kalaska 2014). We used 252 DCs with a wide range of different combinations of Y & B squares; the total number (TE) of B or Y squares in each DC ranged from 0 - 100 of each, as did the net difference in squares (NE). In Task 1, subjects estimated the predominant color of a DC presented in the center of a monitor, and then pointed to either a B or Y target positioned on opposite sides of the DC. In Task 2, a solid B or Y Color Cue in the center of the monitor indicated to which of two DCs on opposite sides of the cue to point; the two DCs had mirror-symmetrical B and Y NE. In Task 3, they saw two monochromatic (either B or Y) DCs with variable numbers of evidence squares and they pointed to the DC with the larger number of squares. While there were some differences in the overall RT durations and error rates in the 3 tasks, subjects’ RTs were poorly related to TE across all tasks. Moreover, unlike many RDM studies, overall performance was better related to the normalized NE in each DC, rather than NE per se. One potential factor is that

RDM direction discrimination is a signal-detection problem (is there a motion bias in the noise?) whereas our task requires an estimate of the relative numbers of easily detected signals (are there more B or Y squares?); judgements of relative numerosity are strongly dependent on normalized evidence (ratio difference of quantities).

Disclosures: **S. Durocher:** None. **J. Milosz:** None. **O. Ierfino:** None. **J. Kalaska:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.15/KK20

Topic: E.04. Voluntary Movements

Title: Mechanisms underlying motor planning under uncertainty

Authors: *L. ALHUSSEIN, R. B. SINGH, M. SMITH
SEAS, Harvard Univ., Cambridge, MA

Abstract: Mounting evidence suggests that uncertainty in tasks requiring goal-directed movements evokes activity in sensorimotor brain areas that reflects multiple possible actions. These actions are thought to indicate distinct motor plans, but the mechanisms by which they are combined into the final observed movement are still unclear. In an experimental setting, the influence of uncertainty on motor planning has been studied in a paradigm where subjects are first presented with two potential targets, and one of them is later cued, but often only after movement onset has occurred. To compensate for this uncertainty, subjects tend to produce intermediate movements between the two targets. Two prominent hypotheses have attempted to provide a mechanistic explanation for how subjects plan these intermediate movements. One idea, the motor averaging hypothesis, states that given multiple potential targets for an action, the uncertain initial motion represents an average of the motor output associated with movements to each potential target. An alternate idea, the optimal planning hypothesis, suggests that the initial motion should instead be planned to optimize for success. This would be accomplished with an initial motion that provides the opportunity for in-flight corrections to be as effective as possible once the final cue is available. A number of previous studies have failed to disambiguate these two hypotheses. To address them, we designed a task for which motor averaging and optimal planning make dramatically different predictions. We used the basic two-target trial paradigm to introduce uncertainty in motor planning. However, we created a dynamic environment that added resistance to specifically increased the effort required for making in-flight corrections to only one of the two potential targets, without interfering inflight corrections to the other target or direct movements to either target. Optimal planning would predict that subjects modify the initial movement to reduce in-flight corrections to the first/resisted target to allow successful corrections against the levied resistance to be feasible. In contrast, the motor averaging

hypothesis would predict that initial movements follow the averaged movement path between the targets, as direct movements to either target do not face interference. While both motor averaging and optimal planning are feasible approaches for motor planning under uncertainty, we hypothesize that the motor system employs optimal planning to maximize task success. The results of this experiment may significantly contribute to the ongoing debate regarding the influence of uncertainty in task goals on motor planning.

Disclosures: L. Alhussein: None. **R.B. Singh:** None. **M. Smith:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.16/KK21

Topic: E.04. Voluntary Movements

Support: CAPES Scholarship

Title: Arm reaching movements are affected by the uncertainty in the targeet location during standing in stroke individuals

Authors: *R. B. GARBUS[1], S. R. ALOUCHE[2], A. G. NARDINI[3], S. M. FREITAS[4]
[1]Univ. Cidade De São Paulo, Santos, Brazil; [2]Univ. Cidade de Sao Paulo, Sao Paulo, Brazil; [3]Univ. Cidade de São Paulo, São Paulo, Brazil; [4]Univ. Cidade De Sao Paulo, Sao Paulo, Brazil

Abstract: Individuals with mild stroke present motor impairments that can affect the performance of arm reaching during standing. Uncertainty in target location has been described to affect these arm movements in healthy individuals and in stroke individuals only in sitting position. Therefore, the aim of this study was to examine the effects of the target uncertainty on the arm reaching during standing in stroke individuals. Nineteen individuals with stroke divided in two groups (9 on the right and 10 on the left side) and eighteen healthy individuals for control group stood in upright position to perform reaching movements with their ipsilateral arm to the brain lesion. Reaches were performed towards a target shown in a monitor and were presented in one of three heights (center, eye level, 10 cm upper or lower to the center target). There were the certain (the participant know where the target was, 60 trials) and uncertain condition (i.e. the target moved for the upper or lower position or remained in the center, 60 trials). The order of trials was randomized.
They were instructed to reach and touch the center of the target as fast as possible after the target changes its color. Time spent from this color stimulus until participants' movement initiation (time of movement onset, TIM), time spent to complete the task (movement time, MT) and accuracy (constant error, CE) were compared among target heights, experimental conditions and groups using Analysis of Variance (ANOVA). For the TIM, ANOVA revealed a condition effect

(p<0.001), individuals spent more time to reach under the uncertainty condition and was 130 ms greater for the stroke group (p<0.001).
For the MT, ANOVA revealed effects of target height (p=0.002) and group (p=0.016).
There was an interaction between height and group (p=0.018), indicating that the effect of height was observed only for stroke group. For the CE, there was an interaction between condition and height (p=0.002) and a triple interaction between condition, height and group (p=0.004). The CE was greater in higher targets for the stroke group and in lower target for control group. The uncertainty in target location affected mainly the TIM for both groups, indicating that the knowledge of the target location is important to plan the reach. The stroke individuals spent more time to initiate their movements and were less accurate for higher targets. The uncertainty in target location affects the arm reaching movements during standing in stroke individuals.

Disclosures: R.B. Garbus: None. **S.R. Alouche:** None. **A.G. Nardini:** None. **S.M. Freitas:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); FAPESP Grant # 2012/19943-0.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.17/KK22

Topic: E.04. Voluntary Movements

Support: NSERC

CFI

FRSQ

Title: Deciding while acting - an investigation of decision-making during ongoing action control

Authors: J. MICHALSKI, *P. E. CISEK
Univ. of Montreal, Montreal, QC, Canada

Abstract: Studies of decision-making, in both psychology and neuroscience, have focused almost exclusively on situations in which subjects must first make a choice and then report it with an action. Such “decide-then-act” paradigms have led to theories in which the brain weighs the costs and benefits of different options, selects one, and then prepares and executes the associated action. However, in our daily lives, we often make decisions while we are already acting (consider almost any sport), and the choice is between an action that is being executed and a new potential action that has become available. Given that the human brain evolved primarily to guide real-time behavior, our theories of decision-making must be able to address decisions made in real-time, during “decide-while-acting” paradigms. The affordance competition

hypothesis (Cisek 2007, Phil. Trans. B) suggests that the brain is continuously processing information about potential actions that are available in the environment, even while performing one of them, and decisions evolve as a competition between these simultaneous action representations. While this hypothesis is supported by a variety of neural and behavioral studies, it has never been tested in scenarios of continuous activity, in which the choice is between continuing one action versus switching to another, as in real-time behavior.
Here, we present an experimental paradigm for studying decision-making during ongoing action control. In the "continuous tracking" task, human subjects use a hand-held cursor to track a target that moves around on the horizontal plane. As long as subjects track the target, they earn rewards (points translated into monetary payments) at a rate that gradually decreases over time and is correlated with the gradually fading luminance of the target. At certain moments, a new target choice is presented somewhere on the screen with a luminance that indicates either a higher or lower reward rate. The subjects can ignore that target and continue tracking the current one, or they can switch to the new target choice, which then starts to move and becomes the tracked target. The placement and timing of appearance of the new target choices is designed to face subjects with decision scenarios that are analogous to situations already well-studied in "decide-then-act" paradigms. For example, we examine how the choice preference curve (with respect to the relative reward value of the new target versus the current tracked target) shifts as a function of the relative biomechanical costs of the tracking movement versus the switching movement, akin to earlier studies in static paradigms (Cos et al. 2011, 2012, 2014, J Neurophysiol).

Disclosures: **J. Michalski:** None. **P.E. Cisek:** None.

Poster

405. The Control of Reaching Movements II

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 405.18/KK23

Topic: E.04. Voluntary Movements

Support: NYS SCIRB C30900GG and C030838GG

NIH 1R01NS092894

Title: Molecular changes in the sensorimotor cortex during learning and recall: Tracking and manipulating PKMzeta

Authors: *J. T. FRANCIS[1], P. GAO[2]
[1]Univ. of Houston, Houston, TX; [2]SUNY Downstate Med. Ctr., Brooklyn, NY

Abstract: Procedural learning and memories, such as those associated with learning how to ride a bike, are believed to be supported by plasticity and reorganization of the sensorimotor cortex. Several studies utilizing a rodent reaching task, as that used in the current work, have shown that procedural learning is accompanied by enhanced synaptic strength and structural modification in the sensorimotor cortex at distinct layers (e.g. layers II/III and V). However, an investigation that causally links these changes with synaptic molecular machinery and behavior has been elusive. This study aims to fill this gap in our current understanding by tracking changes in task performance as well as layer specific modifications in a key molecule, atypical protein kinase M zeta (PKMzeta), that has been shown necessary and sufficient for the maintenance of long-term potentiation (LTP). Our results demonstrate that PKMzata levels decrease in S1 during an early pause in learning on day 3, and peak in S1 and M1 once performance has reached an asymptote on day 9. In addition, continued daily practice after day 9 is accompanied by sustained higher levels of PKMzeta. Past this correlation, we utilized genetic and pharmacological methods to causally perturb PKMzeta during and after learning. We found results indicating the importance of PKMzeta both during learning and in maintaining the procedural memory engram.

Disclosures: **J.T. Francis:** None. **P. Gao:** None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.01/KK24

Topic: E.04. Voluntary Movements

Support: AHA (00035638)

NICHD K12HD055931

5R24HD050821-11

Title: Post-stroke interhemispheric cortical connectivity during active motor states of the paretic and nonparetic limb

Authors: ***J. A. PALMER**[1], L. A. WHEATON[2], P. GURUPRASAD[1], M. R. BORICH[1]
[1]Emory Univ., Atlanta, GA; [2]Biol. Sci., Georgia Inst. of Technol., Atlanta, GA

Abstract: Introduction: Following stroke, atypical interhemispheric (IH) interactions have been observed in stroke survivors with poor motor recovery and may affect post-stroke function. Using synergistic transcranial magnetic stimulation (TMS)-electroencephalography (EEG), our laboratory found differences in IH connectivity between stroke survivors and controls during active but not resting motor states. However, the relationship between IH connectivity and post-stroke strength and impairment of the paretic upper extremity has not been investigated. The

purposes of this study were to (1) investigate the relationship between IH connectivity during paretic (P) and nonparetic (NP) active motor states with P arm motor behavior and, (2) compare the magnitude of IH connectivity in stroke survivors vs. neurologically-intact older adults during P vs. NP muscle activation. **Methods:** Participants with chronic stroke (n=17) and no history of stroke (n=14) completed clinical and neurophysiologic testing using synergistic TMS-EEG. Upper Extremity portion of the Fugl-Meyer (FMUE) assessment and grip strength were assessed. TMS was used to determine M1 resting motor thresholds (RMTs) for the P and NP abductor pollicis brevis (APB). TMS testing was performed during a submaximal contraction of the ipsilateral 1) P and 2) NP APB to evaluate transcallosal inhibition. EEG was recorded during the delivery of 50 suprathreshold (150% RMT) TMS pulses for each condition. IH connectivity was calculated as the post-TMS (0-300ms) imaginary part of coherency (IPC) value between electrodes (C3, C4) within the beta frequency range (15-30Hz). Relationships between IPC vs. FMUE score and P grip strength during each condition were evaluated. We compared IPC in stroke vs. control groups during P vs. non-dominant and NP vs. dominant APB activation. **Results:** We observed a negative relationship between IPC values vs. UEFM ($r=-0.47$, $p=.03$) and P grip strength ($r=-0.48$, $p=.03$) during NP, but not P APB activation. There was a group by condition interaction ($F=5.13$, $p=.03$). In the stroke group, IPC was less during NP ($p=.04$) and greater during P APB activation ($p=.04$) compared to controls. IPC was greater during P vs. NP APB activation ($p=.02$). **Conclusions:** Results indicate that IH connectivity during NP muscle activation was associated with motor behavior poststroke. IH connectivity was abnormal in stroke group during both P and NP muscle activation. Measures of inhibitory neuronal network behavior during NP muscle activation may index salient measures of adaptive corticomotor changes following stroke and potentially offer valuable insights into mechanisms influencing functional recovery.

Disclosures: J.A. Palmer: None. **L.A. Wheaton:** None. **P. Guruprasad:** None. **M.R. Borich:** None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.02/KK25

Topic: E.04. Voluntary Movements

Support: National Science Foundation BCS-145586

Title: Context-dependent brain dynamics during grasping and dexterous manipulation

Authors: *P. MCGURRIN[1], J. FINE[2], K. SCREWS[3], M. SANTELLO[2]
[1]Sch. of Life Sci., [2]Sch. of Biomed. and Hlth. Systems Engin., [3]Sch. for Engin. of Matter, Transport, and Energy, Arizona State Univ., Tempe, AZ

Abstract: Unique sensorimotor processes underlie dexterous manipulation requiring predetermined (constrained) versus self-selected (unconstrained) contact points. The former context involves executing a near-identical motor plan across manipulations via retrieval of previously-learned manipulative forces, whereas the latter requires feedback of digit placement at object contact to drive modulation of digit force distribution to compensate for across-trial variability in digit placement. Our recent work has demonstrated that disrupting the cortical sensorimotor network through non-invasive brain stimulation has differential behavioral effects on constrained versus unconstrained grasping. However, the offline stimulation used in our previous work did not allow us to examine corresponding spatial and temporal features of neural activation. To investigate context-dependent activity of this sensorimotor network, we used electroencephalography (EEG) to quantify spatiotemporal cortical dynamics during unconstrained and constrained grasping. We hypothesized that distinct grasp type-dependent cortical activation and connectivity underlie these grasp contexts due to their unique sensorimotor requirements. Subjects performed a visually-cued grasp and lift task using a precision (thumb-index) grasp, with separate blocks for unconstrained and constrained trials. In all trials subjects were asked to lift an object with an asymmetrical center of mass while minimizing tilt. We combined EEG data with subject's structural MRIs to perform cortical source analysis. Time-frequency analysis of predefined source regions revealed significantly ($p < .01$) greater power in alpha and low beta band power in unconstrained versus unconstrained grasping over left prefrontal, sensorimotor, and parietal regions 400 ms prior to and following the 'go' cue. In addition, Granger connectivity analysis revealed significantly different connectivity from prefrontal and precentral to postcentral regions, as well as parietal to frontal regions. Increases in alpha and beta power have been previously linked to prediction of sensorimotor events. Here we speculate that higher power in these frequency bands is due to increased uncertainty in sensorimotor predictions. Namely, across-trial variability in digit placement during unconstrained grasping necessitates increased reliance on feedback in order to generate correct digit forces to maintain stable performance. These results are the first to establish unique activity of nodes in the fronto-parietal grasping network during tasks requiring differential weighting of feedforward vs. feedback mechanisms.

Disclosures: **P. McGurrin:** None. **J. Fine:** None. **K. Screws:** None. **M. Santello:** None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.03/KK26

Topic: E.04. Voluntary Movements

Support: NIH/NHLBI F32 HL132586-01

NIH/NINDS R01 NS24328

NIH/OD/NINDS P40 ODO10996

NIH/NINDS P30 NS076405

Title: Two representations of the diaphragm in the primary motor cortex

Authors: *L. B. HELOU[1], R. P. DUM[2], P. L. STRICK[3]
[1]Systems Neurosci. Institute, Univ. of Pitt, Pittsburgh, PA; [2]Neurobiology, Ctr. for Neural Basis of Cognition, Systems Neurosci. Inst., Univ. Pittsburgh, Pittsburgh, PA; [3]Neurobio., Systems Neurosci. Institute, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: The diaphragm is a multi-functional muscle. It is the primary muscle for automatic and volitional inspiration. In addition, it is critically involved in vocalization and swallowing, and plays a key role in creating the optimal trans-abdominal pressure gradient for other actions such as posture, micturition, defecation, and vomiting. We used retrograde transneuronal transport of rabies virus from the diaphragm to define the distribution of neurons in M1 that are involved in the control of this multi-functional muscle. We injected rabies virus (CVS-N2c strain, 700-900 uL, 1 $x10^{10}$ pfu) into the costal portion of the diaphragm muscle in cynomolgous monkeys (n=3). We set the survival time (~96 hrs) to be long enough to enable retrograde transneuronal transport of virus to infect neurons in M1. Transport to the cortex required retrograde transport from the muscle to phrenic motoneurons in the spinal cord (1^{st} order neurons); retrograde transneuronal transport from phrenic motoneurons to spinal interneurons and neurons at other sites (e.g., reticular formation) (2^{nd} order neurons); and then another stage of retrograde transneuronal transport from these 2^{nd} order neurons to output neurons in layer V of M1 (3^{rd} order neurons). In all three animals we also observed a small number of labeled neurons in layer III. This indicates that the survival time was long enough to allow transneuronal transport to 4^{th} order neurons. Notably, the labeled neurons in M1 were located in two spatially separate groups. One group was located in the axial body representation of M1 which lies between the arm and leg representations. The second group was located more laterally in the face area of M1. To date, these data represent the most complete picture regarding the neural substrate in M1 that subserves diaphragm motor control in primates. Moreover, the lateral group of neurons appeared to overlap the location of laryngeal representation in M1 that was identified in another study from our laboratory (Cerkevich and Strick, SfN, ’15). Thus, our observations raise the possibility that coordination of laryngeal and breath control during vocalization may be achieved, in part, by co-localizing the cortical control of the two systems in M1.

Disclosures: L.B. Helou: None. **R.P. Dum:** None. **P.L. Strick:** None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.04/KK27

Topic: E.04. Voluntary Movements

Support: Craig Neilsen Foundation 279500

ZIA MH002897

NIH P30 HD018655

NIH P30EY012196

Title: Deconstruction of corticospinal circuits for goal-directed motor skills

Authors: *X. WANG
Neurbiology, Boston Children's Hosp., Boston, MA

Abstract: The corticospinal tract (CST) is the most important voluntary motor control system in humans, but how it transforms cortical commands into accurately executed sequential multi-joint movements in motor skills remains an unresolved issue. By using a retrograde labeling system in mice, we defined the requirement of both total and region-specific CSNs in the accurate execution of sequential movement steps in a skilled forelimb task. In vivo imaging CSN activity during task performance revealed the sequential activation of topographically ordered functional ensembles with moderate local mixing. Region-specific manipulations indicate that CSNs from caudal or rostral forelimb area (CFA or RFA) control reaching or grasping, respectively, and both are required in the transitional pronation step. These region-specific CSNs terminate in different spinal cord levels and locations, therefore preferentially connecting with the premotor neurons of muscles specially engaged in different steps of the skilled motor task. Together, our findings provide a logic for spatially ordered, parallel corticospinal circuits that orchestrate goal-directed motor skills by transforming descending cortical signals into distinct muscle activation patterns.

Disclosures: X. Wang: None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.05/KK28

Topic: E.04. Voluntary Movements

Support: NSERC Grant 418361-2012

Ontario Ministry of Research and Innovation Grant ER14-10-133

Title: Cortical and reticular contributions to response preparation and initiation

Authors: *A. N. CARLSEN[1], D. MASLOVAT[2], N. M. DRUMMOND[1], A. LEGUERRIER[1], J. HAJJ[1], V. SMITH[1]
[1]Univ. of Ottawa, Ottawa, ON, Canada; [2]Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Over the past decade many studies have investigated response preparation through the use of a startling acoustic stimulus (SAS) presented during reaction time (RT) tasks. A common finding is that RTs are dramatically reduced when a startle reflex is induced, often resulting a mean premotor RTs of <70 ms. One hypothesis explaining this "StartReact" response is that details of the motor plan are stored in brainstem centres (e.g. reticular formation) and triggered by startle reflex activation which bypasses normal cortical processing. For example, in patients with hereditary spastic paraplegia with degraded coricospinal pathways, it has been shown that a SAS leads to normalized reaction times, providing support for a subcortically-mediated response pathway. However, other studies involving a SAS have provided evidence that at least some aspects of the prepared response and/or response initiation drive in SAS-speeded RT trials involve contributions from primary motor cortex (M1). For example, when single pulse transcranial magnetic stimulation (TMS) was used to disrupt cortical processing by inducing a cortical silent period, it delayed RTs for both non-SAS and SAS-triggered trials. The present study used a TMS silent period and EMG-EMG coherence analysis to investigate whether different response types involve differential contributions from cortical and reticular centres to the preparation and initiation of movements. In a series of experiments, participants performed targeted single degree of freedom movements with different muscles and task requirements. Experiments 1 and 2 employed TMS to induce a cortical silent period on some trials just prior to initiation of wrist flexion and extension movements. On select trials a SAS unexpectedly preceded the go-signal, whereas on other trials both TMS and a SAS were presented in the same trial. Results from Experiments 1 and 2 showed significant differential RT effects of the TMS between control and SAS trials, as well as between flexion and extension movements in control trials. In Experiments 3 and 4, EMG-EMG coherence was analyzed on control and SAS trials while participants performed RT tasks involving different effectors. Results from these experiments showed significant differences in coherence between movement types in those

frequency bins typically associated with reticulospinal drive. Together the results from these studies suggest that motor actions are represented in a distributed network of cortical and subcortical brain structures, and that the relative contribution of each area depends on the functional and/or anatomical requirements of the task to be performed.

Disclosures: A.N. Carlsen: None. **D. Maslovat:** None. **N.M. Drummond:** None. **A. Leguerrier:** None. **J. Hajj:** None. **V. Smith:** None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.06/KK29

Topic: E.04. Voluntary Movements

Support: CLASS RPG Grant, University of Houston

CAMRI Grant, Baylor College of Medicine

Title: Variability in corticospinal excitability during digit force planning for grasping in humans

Authors: *N. RAO[1], P. J. PARIKH[1,2]
[1]Hlth. and Human Performance, Univ. of Houston, Houston, TX; [2]Sch. of Biol. and Hlth. Systems Engin., Arizona State Univ., Tempe, AZ

Abstract: Variability in motor responses affects the ability to perform motor tasks and learn new motor skills. Motor variability occurs due to the neuromuscular noise during movement execution. Recent work suggests that the central mechanisms during preparation of a motor task may also contribute to motor variability. Specifically, the variability in the firing rate of primate neurons within primary motor (M1) and premotor dorsal cortices during movement preparation contributed to the control of reach trajectory in a motor task. However, how the central mechanisms during movement preparation contribute to motor variability in humans remain unknown. In this study, we sought to determine whether the variability in the corticospinal excitability (CSE) during movement preparation contributes to the control of digit forces during two motor tasks in humans. We delivered single-pulse transcranial magnetic stimulation (TMS) over M1 to assess CSE. Nine healthy young subjects were visually-cued to grip an object at a specific location using the index finger and thumb by exerting either a low grip force (LF; 1N) or a grip force equal to 10% of their maximum voluntary contraction (HF). TMS was delivered at one of the eight latencies from the 'task' cue in a random order: 500, 750, 1000 ('go' cue), 1100, 1200, and 1300 ms. We found a difference in the variability in CSE between LF and HF tasks at 1200 ms after the presentation of the 'task' cue. Interestingly this force magnitude-dependent difference in CSE variability was observed for the first dorsal interosseous and abductor pollicis

brevis muscles, which were directly involved in the exertion of digit force. Similar force magnitude-dependent change in CSE variability was not observed for the abductor digit minimi and flexor carpi radialis (control) muscles. Our findings suggest that the force planning related modulation in the variability of CSE may represent the variability in activity of neuronal population within M1 and/or it may arise from variability in the inputs from neuronal population outside M1. Future work will assess the contribution of M1 and premotor areas to the variability in CSE during the planning of motor tasks.

Disclosures: N. Rao: None. **P.J. Parikh:** None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.07/KK30

Topic: E.04. Voluntary Movements

Title: The relationship between cortical and muscle activity during a motor-cognitive interaction task

Authors: *J. R. LUKOS[1], M. ALAM[2], J. C. BRADFORD[3], D. P. FERRIS[4]
[1]Space and Naval Warfare Systems Center, Pacific, San Diego, CA; [2]Space and Naval Warfare Syst. Ctr. Pacific, San Diego, CA; [3]Army Res. Lab., Aberdeen Proving Ground, MD; [4]Univ. of Michigan, Ann Arbor, MI

Abstract: In contrast to cognitive and motor tasks being done in isolation, performing these tasks simultaneously can affect overall performance. The goal of this study was to better understand the underlying neural processes associated with locomotion under variable cognitive and physical loads. We measured EEG and muscle activity while participants walked on a treadmill with an empty backpack (unloaded condition) or a heavy backpack (40% body weight, loaded condition) at 1 m/s for 1 hour. While walking, subjects also performed a visual oddball task between bouts of isolated walking where 12% of images were targets. We recently found that the EEG revealed differences between target trials during seated, loaded, and unloaded conditions at the electrode level [1] and cortical source level [2], particularly in regions of the brain associated with motor activity. These findings suggests that cortical resources initially dedicated to the cognitive task while seated were reallocated to the motor task while walking. Here, we investigate the effect of changes in physical demand in the muscle activity. We looked at the normalized peak muscle activity for the Tibialis Anterior (TA), Soleus, Rectus Femoris (RF) and Biceps Femoris (BF) muscles during the loaded and unloaded conditions across time for six subjects. We found that the TA and RF exhibited greater muscle activity during the loaded than unloaded condition ($p<0.05$; see Table). Not surprisingly, these preliminary results suggest that walking with a heavy load requires more muscle activity than walking with no load.

Further investigation linking the gait activity to EEG will help to identify which cortical areas contribute to these muscle activity changes.

[1] Bradford et al. (2016). Effect of locomotor demands on cognitive processing. *IEEE EMBC*, Orlando, FL. [2] Lukos et al. (2016). Compensatory neural responses during a physical-cognitive dual task. *SfN*. San Diego, CA.
[3] Benjamini & Hochberg (1995). Controlling the False Discovery Rate: A Practical and Powerful Approach to Multiple Testing. Journal of the Royal Statistical Society.

	Walk 1			Walk 2 + Cog			Walk 3			Walk 4 + Cog			Walk 5		
	Unloaded	Loaded	p-value	Unloaded	Loaded	p-value	Unloaded	Loaded	p-value	Unloaded	Loaded	p-value	Unloaded	Loaded	p-value
TA	0.84 (0.32)	1.3 (0.33)	0.012*	0.92 (0.1)	1.3 (0.38)	0.019*	0.87 (0.12)	1.3 (0.38)	0.012*	0.9 (0.09)	1.31 (0.37)	0.012*	0.88 (0.1)	1.23 (0.38)	0.021*
Soleus	0.96 (0.07)	1.26 (0.34)	0.16	0.79 (0.22)	1.24 (0.46)	0.115	0.82 (0.18)	1.22 (0.46)	0.115	0.87 (0.12)	1.22 (0.42)	0.118	0.94 (0.1)	1.09 (0.34)	0.331
RF	0.73 (0.28)	1.29 (0.56)	0.047*	0.69 (0.32)	1.26 (0.53)	0.02*	0.7 (0.3)	1.26 (0.52)	0.02*	0.72 (0.27)	1.26 (0.53)	0.022*	0.68 (0.36)	1.06 (0.39)	0.022*
BF	0.88 (0.25)	1.08 (0.42)	0.339	0.86 (0.25)	1.13 (0.5)	0.227	0.87 (0.25)	1.14 (0.44)	0.222	0.91 (0.16)	1.18 (0.39)	0.222	0.84 (0.16)	1.15 (0.34)	0.13

Table 1. Mean (and standard deviation) of peak muscle activity between loaded and unloaded conditions. P-values were corrected for multiple comparisons using the Benjamini-Hochberg method [3].

Disclosures: **J.R. Lukos:** None. **M. Alam:** None. **J.C. Bradford:** None. **D.P. Ferris:** None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.08/KK31

Topic: E.04. Voluntary Movements

Support: Swiss National Science Foundation

Title: The role of different motor cortex subregions in goal directed action in mice

Authors: *K. MORANDELL[1], D. HUBER[2]
[1]Univ. of Geneva, Geneve, Switzerland; [2]Basic Neurosci., Univ. of Geneva, Geneva, Switzerland

Abstract: Mammalian motor cortex consists of several interconnected subregions thought to play distinct roles in voluntary forelimb movements, yet their respective role in decision making and motor execution is not clear. Here we performed transient optogenetic inactivations of the caudal forelimb area (CFA) and a rostral forelimb area (RFA) in mice, after they were trained to perform a vibrotactile discrimination task and report their choice by pushing or pulling a joystick after a delay period. We found that choice and motor execution are temporally segregated processes in this behavioral paradigm. CFA and RFA are both necessary during the sensory period for correct choice, while during the delay period neither area alone, but only combined inactivation was able to affect choice. Deficits in movement execution were limited to the execution period and restricted to CFA inactivation. Our findings suggest the existence of transient and partially distributed representations of choice and motor execution across different subregions of motor cortex.

Disclosures: **K. Morandell:** None. **D. Huber:** None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.09/KK32

Topic: E.04. Voluntary Movements

Support: MRC

Title: Modulation of different interneurone networks during proactive and reactive inhibition

Authors: ***V. RAWJI**[1], R. TIBOLD[3], T. FOLTYNIE[2], M. JAHANSHAHI[2], J. C. ROTHWELL[4]
[1]Sobell Dept. of Motor Neurosci. and Movement Disorders, Univ. Col. London, Birmingham, United Kingdom; [2]Sobell Dept. of Motor Neurosci. and Movement Disorders, Univ. Col. London, London, United Kingdom; [3]Sobell Dept. of Motor Neurosci. and Movement Disorders, Univ. Col. London Inst. of Neurol., London, United Kingdom; [4]Inst. Neurol, London, United Kingdom

Abstract: Introduction
Behavioural inhibition is subdivided into reactive (RI) and proactive inhibition (PI), the former becoming active using external cues and the latter an ingrained type of inhibition, serving to facilitate reactive stopping. Recent studies applying Transcranial magnetic stimulation (TMS) in different directions and with specific pulse widths have shown that different inputs into the motor cortex (M1) are differentially modulated depending on behavioural demands. The functional role of these different inputs into M1 with respect to mechanisms of behavioural stopping has not yet been elucidated.
Methods
We employed TMS during either the conditional and normal stop signal task to probe RI and PI in 14 healthy individuals. TMS was applied in two ways: a postero-anterior direction and antero-posterior with 120μs and 30μs pulse widths respectively (PA-120/AP-30). These parameters have been shown to selectively recruit the aforementioned inputs into M1. TMS was applied at different phases of the go (preparation to stop) and stop processes and MEPs recorded from the right FDI muscle, to reflect PI and RI respectively.
Results
During the go trials (PI), PA and AP MEPs were differentially modulated: AP inputs were reduced when PI was implemented, whereas PA inputs were enhanced. This difference reached statistical significance ($p=0.02$, $t=2.464$). For RI we found that stopping the left hand inhibited MEPs more than when the right hand was stopped, for both PA and AP inputs. However, this was statistically significant for PA MEPs only ($p=0.001$, $t=3.941$). Interestingly, we found that the probability of successfully inhibiting increased with the number of preceding go trials, which

was also inversely proportional to PA MEP size.
Discussion
AP and PA inputs into M1 are differentially modulated by behavioural inhibition: preparation to stop specifically inhibits AP inputs whereas stopping is mediated predominantly by PA inputs. We also suggest that the go process is not a global one, and modulates different M1 inputs in varying ways. Finally, we show a physiological predictor of stopping efficacy, and that this too is input specific.

Disclosures: V. Rawji: None. **R. Tibold:** None. **T. Foltynie:** None. **M. Jahanshahi:** None. **J.C. Rothwell:** None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.10/KK33

Topic: E.04. Voluntary Movements

Title: Imitation depends on an intact representation of abstract trajectory shape in dorsal premotor cortex

Authors: *A. L. WONG[1], S. A. JAX[2], L. L. SMITH[2], L. J. BUXBAUM[2], J. W. KRAKAUER[1]
[1]Johns Hopkins Univ. Sch. of Med., Baltimore, MD; [2]Moss Rehabil. Res. Inst., Elkins Park, PA

Abstract: Imitation is an important means of learning skills, but how imitation occurs is not well understood. In part, this is because “imitation” conflates distinct interpretations: achievement of the same task goal, reproduction of the same end posture, and replication of the same kinematic motion. We propose that the latter definition is critical: copying the shape of the trajectory described by the motion of the end-effector in space. In line with this, we recently showed that people generate such trajectory shape representations when planning curved reaches around barriers.
Imitation deficits have been observed bilaterally in patients with apraxia due to left frontoparietal strokes. However, much of this work focused on patients with parietal lesions and actions cued by actors. This has led to two major assumptions about imitation. First, imitation is thought to depend on representing the relative configurations of body parts (body schema); however, bilateral imitation deficits despite unilateral lesions suggest that trajectory shape may be represented in an effector-independent manner. Second, parietal cortex is assumed to perform the computations required for imitation; while this is consistent with fMRI studies of imitation and neurophysiological evidence that activity in parietal cortex modulates with the trajectory taken to hit a target, contrasting neural evidence suggests that neurons in premotor cortex encode movement kinematics of complex trajectories. Thus it is unclear whether trajectory shape is represented in parietal or premotor cortex.

To address these questions, we asked patients with left-hemisphere strokes to imitate movement trajectories. We observed that patients had impaired imitation abilities relative to controls. Deficits were comparable regardless of whether the movement was cued by an actor or a cursor, suggesting an impaired processing stage for representing abstract trajectory shapes in extrinsic space. This could not be attributed to low-level motor impairments: patients imitated point-to-point movements as well as controls. Imitation ability was also uncorrelated to performance when reporting which trajectory shape was observed, suggesting a dissociation between producing trajectory shapes and perceiving stimulus paths. Finally, a voxel-based lesion-symptom mapping analysis found that imitation deficits were associated with lesions in left dorsal premotor cortex (PMd) but not parietal cortex, suggesting that PMd is critical for processing trajectory shapes for movement planning. Together, these findings suggest that imitation critically depends upon the planning of movement shape, which relies upon PMd.

Disclosures: A.L. Wong: None. **S.A. Jax:** None. **L.L. Smith:** None. **L.J. Buxbaum:** None. **J.W. Krakauer:** None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.11/KK34

Topic: E.04. Voluntary Movements

Support: R25N065723

Title: Transfer learning in convolutional neural networks as a tool to analyze animal behavior imaging data

Authors: *A. ARAC[1], S. CARMICHAEL[2], P. GOLSHANI[3]
[1]Neurol., UCLA, Los Angeles, CA; [2]UCLA Sch. Med., Los Angeles, CA; [3]UCLA Dept. of Neurol., Los Angeles, CA

Abstract: To fully understand the principles of motor control or how motor control recovers after brain injuries, motor output must be quantified and analyzed. This approach helps distinguish such effects as compensation vs true recovery and to begin to relate activity in brain circuits to motor output patterns. A key step in such a process is the use of automated animal behavioral experiments. Automated animal behavioral experiments result in large volumes of high quality data. High-speed video-recording is usually used, and an extensive analysis of imaging data, while providing unforeseen insight and information, can be challenging and time-consuming. Thus, there is a huge need for automated analysis methods. The ideal methods would be easily adaptable for different sets of data (object detection, image classification, or segmentation). In addition, these ideal methods would be unbiased, and would require minimal

subjective decision making. Deep learning algorithms such as convolutional neural networks have great promise in automation of image analysis, being used in face detection, object recognition, and self-driving cars. However, one requirement for these networks to achieve high accuracy is the need for large training datasets (millions of images) as the performance of these networks improve significantly when trained with large datasets. This may limit their use in biomedical research given that each type of experimental data has their own training datasets, and creating these training datasets as the ground-truth images can be as challenging. Here, we present use of transfer learning approach. We take an already trained convolutional neural network (i.e. AlexNet, Inception-v3) on ImageNet dataset, and re-train this network with our experimental data. This approach has significant advantages as the already-trained network has variables optimized to detect different features of the images, and good results can be achieved by using relatively small numbers (hundreds to a few thousands) of images from experimental data. We have shown proof-of-concept analysis for paw detection when the mouse is performing a skilled reaching task. To do this, we have created a system where the mouse is head-fixed for future two-photon imaging or electrophysiology experiments, and is performing a reaching task to a food pellet, we image the paw movements with two cameras. We detect the paw by using the convolutional neural networks trained with transfer learning approach, and obtain 2D trajectories. We combine these 2D trajectories to obtain the 3D kinematics of the paw movement. Transfer learning approach can provide an automated, unbiased image analysis of the animal behavior.

Disclosures: **A. Arac:** None. **S. Carmichael:** None. **P. Golshani:** None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.12/KK35

Topic: E.04. Voluntary Movements

Support: NIH 5R01NS025074-24

Katie Samson Foundation

Title: Consistency between task-related neural sub-spaces within and between subjects: Potential for universal neural decoders?

Authors: *C. E. VARGAS-IRWIN[1], J. HYNES[1], J. B. ZIMMERMANN[1,2], J. P. DONOGHUE[2,1]
[1]Dept. of Neurosci., Brown Univ., Providence, RI; [2]Wyss Ctr. For Bio and Neuro Engin., Geneva, Switzerland

Abstract: The activity of networks of neurons addressing similar computational challenges should reflect common information processing motifs. We hypothesize that for motor cortical areas driving upper limb motion, the common structure of the end effector will result in encoding features that remain consistent between subjects. In order to test this hypothesis, we use spike train similarity space (SSIMS) analysis to generate low dimensional embeddings that capture the intrinsic relationship between network states. Comparing these state spaces allows us to quantify the similarity between encoding schemes across different networks. In the present work we compare SSIMS maps generated using activity in primate primary motor cortex of monkeys performing a planar center out reaching task. The resulting state spaces consisted of eight clusters associated with different reaching directions. Instead of being distributed in a uniform circle, the clusters were arranged in a more complex asymmetric shape that was consistently observed across different datasets. We demonstrate that using simple normalization techniques and matching the directions of maximum variance, it is possible to combine neural state spaces from different sessions and even different subjects into one cohesive whole. This can be accomplished using the intrinsic properties in each neural dataset and with no a priori knowledge of the different trial conditions. The resulting combined space can be used as a basis for decoding models applicable across sessions and subjects, demonstrating a remarkable degree of convergence between the neural state spaces analyzed. Our results show that SSIMS captures intrinsic features of motor control processes applicable to multiple subjects performing the same task. This finding suggest that it should be possible to train 'universal' neural decoders that can be applied to new subjects out of the box.

Disclosures: C.E. Vargas-Irwin: None. **J. Hynes:** None. **J.B. Zimmermann:** None. **J.P. Donoghue:** None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.13/KK36

Topic: E.04. Voluntary Movements

Support: NIH Grant 5R21NS094946-02

Title: Motor learning for corticospinal and corticobulbar pathways

Authors: *S. PARK[1], A. CASAMENTO-MORAN[1], M. L. SINGER[2], A. E. ERNSTER[2], B. YACOUBI[1], I. A. HUMBERT[2], E. A. CHRISTOU[1]
[1]Applied Physiol. and Kinesiology, [2]Speech, Language, and Hearing Sci., Univ. of Florida, Gainesville, FL

Abstract: The use of visual feedback is critical for motor learning. It remains unknown whether the use of visual feedback for motor learning differs for effectors controlled by different descending pathways. Here, we compare motor learning of the ankle joint and tongue because they represent the corticospinal and corticobulbar pathways, respectively. Twelve young adults (19.63 ± 2.11 years, 6 females) practiced a tracking task (combination of four frequencies at 0.02, 0.37, 0.5, and 1 Hz) with ankle dorsiflexion and with tongue elevation for 100 trials. The participants practiced each effector (ankle or tongue) in different days and the order of the effector was counterbalanced. Following practice, participants performed the same tracking task with simultaneous contractions of the tongue and ankle (dual tracking task; transfer) with three different visual feedback conditions (no visual feedback, visual feedback only for ankle, visual feedback only for tongue). We recorded the force output from the tongue and ankle and quantified the force accuracy (RMSE) from each effector. When there was no visual feedback of the motor output or target (testing memory of the practiced task), the performance of the dual task was similar for the ankle and tongue. In contrast, the RMSE was lower for the ankle than the tongue regardless whether participants received visual feedback about the ankle or tongue ($P <$ 0.02). These results provide novel evidence that learning a motor task that requires integration of visual feedback is better for the corticospinal pathway (ankle joint) than the corticobulbar pathway (tongue).

Disclosures: S. Park: None. **A. Casamento-Moran:** None. **M.L. Singer:** None. **A.E. Ernster:** None. **B. Yacoubi:** None. **I.A. Humbert:** None. **E.A. Christou:** None.

Poster

406. Cortical Planning and Execution: Neural Correlates of Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 406.14/LL1

Topic: D.07. Vision

Support: Grant-in-Aid for JSPS Research Fellow

Title: Effects of pseudoexperience on the understanding of hemiplegic movements: An fMRI study by physical therapists as subjects

Authors: *R. WATANABE, N. KATSUYAMA, N. USUI, M. TAIRA
Cognitive neurobiology, Tokyo Med. and Dent. Univ., Bunkyo-ku Sendagi, Japan

Abstract: The fronto-parietal cortical network is suggested to play a fundamental role in the understanding of observed actions (action observation network, AON).
Previous studies have shown that AON is more activated in the observer with actual experience of observed action than without. However, it is not known whether the pseudoexperience of observed movement also have the same effect on the AON as the actual experience. Physical

therapists (PTs) routinely observe the hemiplegic movements of stroke patients in clinic. Thus, they are thought to have pseudoexperience of the hemiplegic movements in the treatment, such as careful touching and observing of the hemiplegic bodies, even though almost none of them experienced hemiplegia.
In the present study, in order to understand the effect of pseudoexperience, we measured cortical activity of 19 PTs and 19 non-PTs using functional MRI (fMRI). Two conditions were prepared, such as observing movies of the stroke patients clasping and unclasping 1) with the hemiplegic hand and 2) with the non-hemiplegic hand. Immediately after the fMRI scan, participants again observed all the movies and were asked to assess the patients feeling to their own hemiplegic movements by the questionnaire. In fMRI measurements, the AON in the PTs was more activated than in the non-PTs during observing the
hemiplegic movements. Furthermore, the anterior/middle cingulate cortex (aMCC) which is thought to relate to negative affect was also more active, and the left aMCC increased its connectivity to the right supramarginal gyrus which is thought to relate to body representation, in psychophysiological interaction analysis. In addition, behavioral test indicated that the PTs assessed more accurately patients' feelings of difficulty associated with hemiplegic movements than the non-PTs. These findings suggest that even the pseudoexperience also have the same effect on the AON as the actual experience, and may help for better understanding of the observed movements through activation of the AON. Furthermore, the better understanding of difficulty of hemiplegic movements, the pseudoexperience may affect limbic system through modulating own body representations.

Disclosures: R. Watanabe: None. **N. Katsuyama:** None. **N. Usui:** None. **M. Taira:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.01/DP09/LL2 (Dynamic Poster)

Topic: E.04. Voluntary Movements

Support: Barrow Neurological Foundation

A group of private donors

Title: An open database of cat neuronal activity

Authors: I. A. NALL[1], I. BRAVERMAN[1], J. T. GOODROAD[1], F. FLORES[1], F. JOHNSON[1], A. RIVARD[1], *I. N. BELOOZEROVA[2]
[1]Arizona State Univ., Tempe, AZ; [2]Neurobio., Barrow Neurolog. Inst., Phoenix, AZ

Abstract: The cat is one of the best researched higher vertebrates. It is a model animal for studies of several common diseases. Except for the frontal lobes, cat brain is quite similar to the brain of primates. It is well studied but yet little understood. To promote understanding of the brain in general, we have created an open database of cat brain neuronal activity during various behaviors to allow all interested parties to search and analyze available cat brain data.
We formed a back-end system that utilizes the existing front-end functionality of WordPress, an open-source content management system, which is based on PHP scripting language and also uses an attached SQL relational database management system. We have merged that SQL with our custom MySQL database to act as our main data repository. With our interface and general functions, such as file-uploading, being handled by WordPress, we focused on modifying the system to make it better tailored to our requirements. We created a series of custom plugins, themes, scripts and widgets to allow for: (i) unique multi-data-point searching first through "AND" and then "OR", (ii) varied searches that use both user-created and system-generated tags; (iii) increased data variety and consistency, with full data manipulation; and (iv) an administration layer, which allows admins to approve and monitor users.
In this dynamic poster, we will demonstrate the central features of the database: general searching, easy access to data, and data management. We will perform sample searches in response to attendees' requests. By the time of the meeting, the database will contain ~500 GB of data on single neuron activity in the cat motor cortex and motor thalamus during overground locomotion with and without obstacles. Both raw and processed records will be available. Neuron identification, including data on axonal projections and conduction properties obtained in live subjects will be available. Approximate location of cell bodies within the structure determined by histology will be available. All data will be accompanied with behavioral data on task performance.
The cat brain database is started with data on locomotion collected in the laboratory of Dr. Beloozerova since 2000 as a part of NIH funded research. Later, data on cat brain activity during treadmill locomotion, balancing, and scratching will be added. Three-dimensional kinematic data on all behaviors will be added. The database is open to other laboratories for depositing their data on cat brain activity and body biomechanics. By making the data publicly available we seek to facilitate further research of the cat brain, which ultimately will lead to better understanding of human brain.

Disclosures: I.A. Nall: None. **I. Braverman:** None. **J.T. Goodroad:** None. **F. Flores:** None. **F. Johnson:** None. **A. Rivard:** None. **I.N. Beloozerova:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.02/LL3

Topic: E.04. Voluntary Movements

Support: P.D.G. was supported by the European Union's Horizon 2020 Research and Innovation Programme under Grant Agreement No. 720270 (HBP SGA1)

Title: A 'landscape view' of motor planning

Authors: *P. DEL GIUDICE[1], G. BAGLIETTO[2], S. FERRAINA[3]
[1]Italian Natl. Inst. of Hlth., Rome, Italy; [2]IFLYSIB, Inst. de Fisica de Liquidos y Sistemas Biologicos (UNLP-CONICET),, La Plata, Argentina; [3]Dept Physiol. and Pharmacol., Sapienza Univ. Rome, Rome, Italy

Abstract: We recently described (Baglietto et al, Plos One 2017) an approach to the analysis of multi-dimensional time series, like those obtained from multiple simultaneous electrophysiological recordings, based on a density-based clustering (DBC), and showed in a modeling study its potential for efficient representation, inference and complexity estimates. In this contribution we used DBC to analyze the spatio-temporal organization of cortical activity patterns during the execution of a 'countermanding' task, in which a monkey must perform a reaching movement to a target on a screen ('no-stop trials'), unless a stop signal prescribes to withhold the movement ('stop-trials'); no-stop and stop trials are randomly intermixed, and the stop signal occurs at random times within the monkey's reaction time. In a previous study using the same task (Mattia et al, J. Neurosci. 2013), a spectral analysis of multi-unit activity (MUA) from single electrode recordings of the dorsal premotor cortex (PMD) suggested that the development of the motor plan proceeds as a cascade of sudden UP-DOWN/DOWN-UP transitions in the local cortical neural population (module). The analysis suggested that transitions are autonomously generated in most excitable modules, and propagate to less excitable ones. In their relation with the completion of the motor plan, the time patterns of transitions were predictive of the movement time in no-stop trials, and of the correct or incorrect behavior in stop trials. Here we present preliminary result of the analysis of simultaneous MUA recordings from a 96-electrodes grid in PMD during the same countermanding task. We perform DBC on the 96-dimensional MUA time series, allowing to code it as a discrete sequence of clusters' centroid labels (so each label represents a 96-elements vector of MUA values in a given time bin, which is the centroid of the cluster currently visited by the neural dynamics). This coding offers a clean and compact view of the spatio-temporal dynamics, which confirms the picture suggested by the previous analysis and offers an explicit representation of the task-related activation sequences of cortical modules. Besides, to inquire into long memory effects in the dynamics, from the sequence of centroid labels we first estimated the matrix of transition probabilities between clusters, from which we constructed surrogate Markov centroid sequences. For both real and surrogate sequences we computed the Lempel-Ziv complexity, and defined an index measuring their relative complexities, thereby obtaining a quantitative measure of long-memory, non-Markov dynamic components.

Disclosures: P. Del Giudice: None. **G. Baglietto:** None. **S. Ferraina:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.03/LL4

Topic: E.04. Voluntary Movements

Support: NIH R01 NS24328 (PLS)

NINDS P30 NS076405 (PLS)

Title: Inhibition of protein synthesis in M1 of monkeys disrupts performance of sequential movements guided by memory

Authors: *M. OHBAYASHI[1,2,3,4], P. L. STRICK[1,2,3,4]
[1]Univ. of Pittsburgh, Sch. of Medicine, Syst. Neurosci. Inst., Pittsburgh, PA; [2]CNBC, Pittsburgh, PA; [3]Dept. of Neurobio., Pittsburgh, PA; [4]Brain Inst., Pittsburgh, PA

Abstract: Motor skills may be improved with extensive practice to attain expert performance (e.g., playing the violin). Recent studies in monkeys indicate that extended practice on a motor skill alters patterns of neural activity and functional activation in M1 (Matsuzaka et al., '07; Picard et al., '13). We previously showed that inhibition of protein synthesis in M1 of monkeys disrupted performance of sequencial movements guided by memory, but not performance of visually-guided reaching (Ohbayashi et al., SFN Abstr., '15). These and other results suggest that M1 is a site of storage for highly practiced movements. Here we examine the effect of inhibiting protein synthesis in M1 on the development of expertise during a sequential motor task.We trained 2 monkeys (Cebus Apella) to make sequential reaching movements in two tasks (Ohbayashi et al., '16). In the Random task, movements were guided by vision. New targets were shown using a pseudorandom sequence 100 ms after contact of the prior target. In the Repeating task, movements were internally guided. New targets were shown according to a repeating 3 element sequence (e.g., 5-3-1-5-3-1 ...) 400 ms after contact of the prior target. The animal was allowed to contact the next target in the sequence before it was shown. The longer delay between target contact and the appearance of the next target promoted the occurrence of predictive responses. With practice, monkeys could perform the Repeating task without any visual cues. After the monkeys became proficient in the performance of the 1st trained sequence, a 2nd sequence (e.g., 1-2-4-1-2-4...) was introduced. The monkeys performed the two sequences and the Random task in alternate blocks.After training for more than 200 days on the 1st sequence and 50 days on the 2nd sequence, we injected the protein synthesis inhibitor anisomycin (5 µl of 100 mg/ml) at 2 sites in the arm area of M1 (monkey 1, n = 5; monkey 2, n = 1). After the anisomycin injections, Response Time (RT) and the number of errors increased significantly only during performance of the Repeating task. We compared the effects of anisomycin injections on performance of the 1st and 2nd sequences in each session. The RT increase induced

by the anisomycin injection was greater for the 2nd sequence than for the 1st one (n = 6). These results confirm the involvement of M1 is the development of expertise through practice. In addition, they suggest that the more recently learned 2nd sequence is more vulnerable to the inhibition of protein synthesis than the older 1st sequence.

Disclosures: M. Ohbayashi: None. **P.L. Strick:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.04/LL5

Topic: E.04. Voluntary Movements

Support: NIH Grant NS079471

Institutional Funds for Startup

Title: Intrinsic connections of motor cortex columns revealed with intracortical microstimulation and optical imaging in squirrel monkeys

Authors: N. S. CARD[1], A. SLOAN[1], *O. A. GHARBAWIE[2]
[2]Dept. of Neurobio. & Systems Neurosci. Inst., [1]Univ. of Pittsburgh, Pittsburgh, PA

Abstract: In primary motor cortex (M1), a roughly concentric topography exists for the motor representations of the hand, elbow, and shoulder. Cortical columns within these representations send corticospinal projections that can influence activity in groups of arm and hand muscles. The muscle synergies needed for manual movements are therefore predicated on coordinated activity between M1 columns within the arm and hand representations. Multiple communication channels have the potential to coordinate activity in M1 columns. Intrinsic M1 connections represent the most direct channel of communication between M1 columns, but are perhaps the least understood among M1 connections. The objective of the present study is to investigate the spatial organization of the intrinsic connections of M1 in columns within the arm and hand representations. In three squirrel monkeys, we focused on mediolateral rows of cortical columns in M1, wherein motor output changes but other defining features are invariant. To study an individual column, we first determined the output targets of that column via intracortical microstimulation (ICMS) and electromyographic (EMG) recordings. Second, we identified zones in M1 that are connected to that column using ICMS (trains of 150 pulses, 0.2 ms/pulse, 300 Hz, 1000 μm below pia) and concurrent intrinsic signal optical imaging (630 nm illumination). For all M1 columns investigated in this study, the most prominent activation was a spatial cluster (~ 2.0 mm^2) of contiguous columns that surrounded the stimulating microelectrode. In addition, columns were preferentially connected with other clusters (~ 0.5 mm^2) of columns. The muscle

targets of the connected columns overlapped with the muscle targets of the microstimulation site. Our results build on tracer studies that showed that the *anatomical* connections of the thumb representation in monkeys (Huntly and Jones, 1991) and the wrist representation in cats (Keller, 1993), are widely distributed across the entire forelimb representation. Here, we show that the *functional* connections of M1 columns within the forelimb representation are spatially biased towards M1 columns that target the same muscle groups. Thus, our results to date suggest that *functional* connections within M1 may be primarily concerned with coordinating matched columns. In this framework, other communication channels (e.g. thalamic inputs, intra-areal connections) may be responsible for coordinating non-matched M1 columns. We are currently testing the effects of ICMS in one column on the activity of single units in connected columns to determine if interactions between connected columns are excitatory or inhibitory.

Disclosures: N.S. Card: None. **A. Sloan:** None. **O.A. Gharbawie:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.05/LL6

Topic: E.04. Voluntary Movements

Support: JSPS KAKENHI 16K17369

JSPS KAKENHI 22830086

Title: Area-specific involvement of frontal areas and the basal ganglia in goal-directed behavior in monkeys

Authors: *Y. NAKAYAMA[1,2,3], T. YAMAGATA[2,3], N. ARIMURA[2,3], E. HOSHI[1,2,3]
[1]Neural Prosthesis Project, [2]Frontal Lobe Function Project, Tokyo Metropolitan Inst. of Med. Sci., Tokyo, Japan; [3]Tamagawa Univ. Brain Sci. Inst., Tokyo, Japan

Abstract: Regions in the frontal lobe form closed loop circuits with the basal ganglia. The areas composing these circuits are considered to play a role in achieving goal-directed motor behavior. However, the specific roles that each area composing these circuits plays still remain elusive. In the present study, we examined neuronal activity in multiple brain areas of monkeys (*Macaca fuscata*) while they performed a conditional visuo-goal task designed to include separate processes for determining a behavioral goal (reaching towards a right or left potential target) on the basis of visual object instructions, specifying actions (direction of reaching) to be performed on the basis of the goal, and preparing and executing the action (Nakayama et al., 2008, 2016). We compared neuronal activity in eight areas of the frontal cortex and basal ganglia: the ventrolateral and dorsolateral prefrontal cortex (vlPFC and dlPFC, respectively), pre-dorsal

premotor area (pre-PMd, or area F7), dorsal premotor area (PMd, or area F2), ventral premotor area (PMv), caudal cingulate motor area (CMAc), primary motor cortex (M1), and globus pallidus (GP). We found activity of neurons selective for (1) visual features of the instruction cues, (2) goal determination, (3) action specification, and (4) preparation and execution of the action. We next applied k-means clustering method to classify neurons into several groups based on patterns of their activity. We found clusters of neurons that were active when the monkeys determined a goal, specified an action, prepared the action, or executed the action. The proportions of neurons that belonged to each cluster were different between the eight areas. Based on these analyses, we found that (1) neurons in the vlPFC amply encoded visual features of objects, (2) neurons involved in the processes of determining goals and specifying and maintaining actions were found mainly in the vlPFC, dlPFC, pre-PMd, and PMd, (3) neurons involved in action preparation and execution were mainly found in the PMd, PMv, and M1, (4) neurons in the CMAc were involved in action execution regardless of directions of the action, and (5) neurons in the GP were involved in goal determination and action specification, but they did not maintain the information. These results suggest that each frontal area coordinates with the basal ganglia, and these areas contribute to goal-directed behavior in an area-specific manner.

Disclosures: **Y. Nakayama:** None. **T. Yamagata:** None. **N. Arimura:** None. **E. Hoshi:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.06/LL7

Topic: E.04. Voluntary Movements

Support: NINDS R24NS098536

NINDS R01NS089679

Title: Premotor network exploration and reinforcement during practice of a stereotyped, learned motor action

Authors: *W. A. LIBERTI, III[1], J. SHEN[1], D. P. LEMAN[1], N. PERKINS[2], T. J. GARDNER[1]
[1]Biol., [2]Boston Univ., Boston, MA

Abstract: With practice, learned motor actions can be maintained for decades, but the biological basis of skill acquisition and persistence remains largely unknown. We do know that precise motor skills are learned and maintained through practice: a process of exploratory trial-and-error learning where actions are evaluated and ‘good’ performances are reinforced. For example, the adult male zebra finch sings a highly stereotyped courtship song that is stable for years. These finches will sing ‘undirected’ in isolation, and these songs are less stereotyped than when their

performance is ‘directed’ to another finch. In parallel, neurons impinging onto motor cortical area RA show increased variability in their firing patterns during undirected singing, and this variability is thought to provide a substrate for vocal exploration and trial and error learning. We deploy head mounted miniature microscopes and cell-type specific genetic tools in the pre-motor nucleus HVC, one synapse upstream of RA. We observe and compare the underlying neural activity during motor production of both ‘undirected’ and ‘directed’ song, to determine if this area also undergoes state-dependent changes in variability. On average, individual neurons have similar firing times in both conditions. However, preliminary data suggests that during undirected practice, individual neurons independently explore different firing modes. In contrast, when song is directed to a female, this exploration is largely diminished, and variance becomes more correlated across the network. This suggests that during undirected singing, neurons in HVC may also express variations that could form a substrate for trial and error learning to minimize vocal errors. In this poster we examine the correlation structure of neural variations in greater detail.

Disclosures: W.A. Liberti: None. **J. Shen:** None. **D.P. Leman:** None. **N. Perkins:** None. **T.J. Gardner:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.07/LL8

Topic: E.04. Voluntary Movements

Support: SA 2575/3-1

JST PRESTO

DFG EXC 307

DFG SA 2575/2-1

MEXT/JSPS KAKENHI 26119536

MEXT/JSPS KAKENHI 26242088

MEXT/JSPS KAKENHI 15H05710

Title: Selective suppression of local circuits during movement preparation in the mouse motor cortex

Authors: *T. R. SATO[1], M. HASEGAWA[2], K. MAJIMA[3], T. ITOKAZU[5], Y. KAMITANI[4], T. SATO[6]

[1]Ctr. For Integrative Neurosci., Tuebingen, Germany; [2]Univ. of Tuebingen, Tuebingen, Germany; [3]Grad. Sch. of Informatics, Kyoto Univ., Kyoto-Shi, Japan; [4]Grad. Sch. of Informatics, Kyoto Univ., Kyoto, Japan; [5]Osaka Univ., Osaka, Japan; [6]Technische Univ. München, Muenchen, Germany

Abstract: Prepared movements are more efficient than those that are not prepared for. Although changes in cortical activity have been observed prior to a forthcoming action, the circuits involved in motor preparation remain unclear. Here, we use in vivo two-photon calcium imaging to uncover changes in the motor cortex during variable waiting periods prior to a forepaw reaching task in mice. Consistent with previous reports, we observed a subset of neurons with increased activity during the waiting period; however, these neurons did not account for the degree of preparation as defined by reaction time (RT). Instead, the suppression of activity of distinct neurons in the same cortical area better accounts for RT. This suppression of neural activity resulted in a distinct and reproducible pattern when mice were well prepared. Thus, the selective suppression of network activity in the motor cortex may be a key feature of prepared movements.

Disclosures: T.R. Sato: None. **M. Hasegawa:** None. **K. Majima:** None. **T. Itokazu:** None. **Y. Kamitani:** None. **T. Sato:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.08/LL9

Topic: E.04. Voluntary Movements

Support: NIH grant # NS061963

Title: Thalamo-cortico-thalamic circuits in a premotor-like frontal cortical area in the mouse

Authors: *K. GUO, N. YAMAWAKI, G. M. SHEPHERD
Physiol., Northwestern Univ., Chicago, IL

Abstract: What is the cellular basis for communication between thalamus and cortex? This question has been most extensively investigated in sensory systems, leading to concepts of thalamo-cortico-thalamic (T-C-T) loops, which particularly involve thalamocortical (TC) neurons in the thalamus and layer 6 corticothalamic (CT) neurons in the cortex. However, our recent studies indicate a distinct organization for T-C-T circuits in primary motor cortex (M1) of the mouse (Yamawaki & Shepherd, 2015, J Neurosci), including a paucity of monosynaptic connections between TC neurons in the ventrolateral nucleus and CT neurons in M1, in either direction. Instead, T-C-T loops of M1 appear to be polysynaptic, involving multiple nuclei in the

thalamus and multiple projection classes in the cortex, including pyramidal tract (PT) and intratelencephalic (IT) neurons in addition to CT neurons. Here, we extended our studies of motor-related T-C-T circuits to assess cellular connections between higher-order motor areas in the anterior frontal cortex and higher-order nuclei in motor thalamus. We applied optogenetic-electrophysiology tools to determine how IT, PT, CT, and TC neurons participate in the cell-type-specific connections that form T-C-T circuits. We focused on the anterior lateral motor (ALM) area, a frontal cortical region with premotor-like properties (Li et al., 2015, 2016, Nature), and its connections with the ventromedial (VM) nucleus, a basal-ganglia-recipient region of motor thalamus. The findings show several unusual properties of T-C-T circuits in this system, differing from both sensory and M1-related T-C-T circuits. In the T-->C direction, these included strong excitation from VM-TC axons onto layer 1 apical tuft dendrites of PT neurons that were identified by retrograde labeling as projecting back to VM. In the C-->T direction, PT axons strongly excited TC neurons in VM that were identified as projecting back to ALM. Thus, the results provide evidence for direct, recurrent loops in this higher-order motor-frontal T-C-T system in the mouse, mediated by PT neurons in ALM and TC neurons in VM. We speculate that such recurrence could be important for supporting reverberant activity associated with motor planning.

Disclosures: K. Guo: None. **N. Yamawaki:** None. **G.M. Shepherd:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.09/LL10

Topic: E.04. Voluntary Movements

Support: NBRC Core Funds

Title: Topography in the mouse motor cortex is a mosaic

Authors: *N. JAIN, S. R. JOHN, P. HALDER
Natl. Brain Res. Ctr., Manesar, India

Abstract: Mammalian motor cortex is considered to be globally topographic and locally non-topographic. Although the neurons that control movements of different body parts are topographically organized in the motor cortex, within the representation of a body part the organization is not precisely topographic. Such movement maps in the motor cortex have been described for a number of mammalian species including rodents and non-human primates. Topographic organization of the motor cortex is usually determined by intracortical microstimulation (ICMS). ICMS involves using microelectrodes to inject small amounts of current to stimulate layer 5 neurons. The stimulating current is reduced to the smallest threshold

value that elicits a visible movement of a body part. Here we used ICMS to determine organization of the motor cortex in mice.
We found that although there is an overall topographic organization in the mouse motor cortex, the locations of boundaries of representations of various body parts are variable. There is a medial whisker movement area, bordered on the lateral side by the forelimb area. Caudally there are small trunk and hindlimb movement regions, and a rather large tail area. Within these representations, there are scattered patches of neurons from where movements of other body parts, particularly the tail could be evoked.
Interestingly, stimulation at currents that were slightly higher than the threshold currents (i.e. suprathreshold currents) evoked movements of additional body parts at nearly all the sites. The increase in threshold currents required for additional movements was generally as low as 1 µA. For example, at suprathreshold currents whisker movements could be evoked from the forelimb area and *vice versa*. The number of tail movement sites increased throughout the motor cortex. The additional movements were unlikely to be due to the point-spread of the current, because these movements were observed not just at the boundaries of the representations, but throughout the motor cortex. Moreover, the movements evoked at suprathreshold currents could be of non-adjacent and distant body parts. In rats, in contrast, suprathreshold currents do not evoke movements of additional body parts, except that movements of the neck muscles are evoked from the whisker area (Tandon et al., Eur J Neurosci, 27:228, 2008).
We conclude that the mouse motor cortex has a mosaic representation of movements of different body parts.

Disclosures: **N. Jain:** A. Employment/Salary (full or part-time):; National Brain Research Centre. **S.R. John:** A. Employment/Salary (full or part-time):; National Brain Research Centre. **P. Halder:** A. Employment/Salary (full or part-time):; National Brain Research Centre\.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.10/LL11

Topic: E.04. Voluntary Movements

Support: NIH grant NS061963

NIH grant EB017695

Title: Corticocortical signaling drives activity in a downstream area efficiently, reliably, reversibly, and scalably

Authors: *X. LI[1], N. YAMAWAKI[1], K. P. KORDING[2], G. M. G. SHEPHERD[1]
[1]Physiol., [2]Physical Med. and Rehabil. and Physiol., Northwestern Univ., Chicago, IL

Abstract: How effectively does activity in an upstream cortical area drive activity in a downstream area? Optogenetic photostimulation and multi-unit electrophysiology were used to characterize evoked upstream and downstream spiking activity in a parietofrontal pathway linking retrosplenial cortex (RSC) to posterior secondary motor cortex (M2) in mice, a pathway recently characterized at the level of cell-type-specific corticocortical connectivity (Yamawaki et al., 2016, J Neurosci). RSC neurons including their axons projecting to M2 were labeled with channelrhodopsin-2 (ChR2) by injection of AAV in RSC in wild-type mice. Optical fibers were placed over RSC, to activate the upstream RSC neurons directly, and also in M2, to activate the axons of the same RSC neurons and thereby activate them antidromically. Activity was simultaneously recorded in both RSC and M2 using linear arrays (silicon probes) with 32 channels each, to sample multi-unit activity across cortical layers in both areas. Photostimuli delivered to the upstream area (RSC) were robustly converted into local activity, which in turn drove activity in downstream M2 with nearly the same efficiency and reliability. Reverse (antidromic) driving, delivering photostimuli to M2, was similarly robust. In either direction, downstream activity scaled approximately linearly with stimulus intensity and sub-linearly with duration, and with an overall log-like relationship in this input-output system. Shorter-duration higher-intensity inputs were most effective, consistent with an adaptation-like process. The results show that corticocortical signaling in this pathway exhibits efficiency, reliability, reversibility, and scalability. The findings suggest that corticocortical signaling supports inter-areal communication across a wide dynamic range of activity levels and temporal patterns, but preferentially for briefer bursts of activity.

Disclosures: X. Li: None. **N. Yamawaki:** None. **K.P. Kording:** None. **G.M.G. Shepherd:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.11/LL12

Topic: E.04. Voluntary Movements

Support: European Research Council (ERC-2010-StG-260590, ERC-2015-CoG-682422)

Deutsche Forschungsgemeinschaft (DFG, Exc 257 NeuroCure, FOR 1341, FOR 2143)

Berlin Institute of Health (BIH)

Thyssen Foundation

Helmholtz Society

Title: Parvalbumin expressing GABA-ergic neurons in primary motor cortex signal reaching

Authors: *D. HOFFMANN[1,2], L. ESTEBANEZ[1,2,3], B. C. VOIGT[1,2], J. F. POULET[1,2]
[1]Max-Delbrück Ctr. For Mol. Med. (MDC), Berlin, Germany; [2]Neurosci. Res. Ctr., Cluster of Excellence NeuroCure/ Charité-Universitätsmedizin, Berlin, Germany; [3]Unité de Neurosciences, Information et Complexité, UNIC-FRE3693, Ctr. Natl. de la Recherche Scientifique, Gif-sur-Yvette, France

Abstract: The control of targeted reaching is thought to be shaped by distinct subtypes of local GABA-ergic inhibitory neurons in primary forelimb motor cortex (M1). However, little is known about their action potential firing dynamics during reaching. To address this, we recorded the activity of parvalbumin-expressing GABA-ergic neurons, fast spiking units and regular spiking, presumed excitatory, units in layer 5 of the mouse forelimb M1 during a sensory triggered reaching task. An acoustic cue signaled the onset of a trial, then mice reach and press a sensor following a brief vibrotactile trigger stimulus delivered to the same paw. M1 activity is required for the task, as injection of the GABA-A agonist muscimol disrupted reaching. Parvalbumin-expressing neurons showed short latency responses to the acoustic cue and vibrotactile trigger input as well as an increase in firing at reaching onset that was positively correlated with the amplitude of reaching. Unexpectedly, parvalbumin-expressing neurons fired before regular spiking units at reach onset and maintained high firing rates until the end of the reach. Thus, increasing M1 parvalbumin-expressing neuron firing rates may play a role in the initiation of voluntary reaching.

Disclosures: D. Hoffmann: None. **L. Estebanez:** None. **B.C. Voigt:** None. **J.F. Poulet:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.12/LL13

Topic: E.04. Voluntary Movements

Support: NIH / NINDS grant # NS061963

Title: Transcranial laser scanning photostimulation and video motion tracking for optogenetic cortical motor mapping of mouse corticospinal neurons

Authors: *L. LAMBOT, J. M. BARRETT, X. LI, G. M. SHEPHERD
Dept. of Physiology, Feinberg Sch. of Med., Northwestern Univ., Chicago, IL

Abstract: Motor mapping, based on stimulating different cortical locations and measuring the evoked movement, has long been used to define and study motor cortical areas. Traditional electrophysiological mapping methods have several limitations, including lack of cell type specificity and poor spatial resolution. Light-based mapping (LBM; Ayling et al., 2009) has

emerged as a method to overcome both these limitations, and several groups have recently developed various implementations of this technique for investigating limb and whisker movements in mice. We will present progress towards developing a variant of LBM based on combining transcranial laser scanning photostimulation (tcLSPS) with video motion tracking (VMT) using inexpensive cameras, with the aim of using tcLSPS-VMT to map the cortically evoked movements generated by selective photostimulation of channelrhodopsin-2 (ChR2) expressing corticospinal neurons. To selectively label corticospinal neurons with ChR2, we injected the cervical spinal cord with rAAV2-retro-Syn-ChR2-GFP (gift of Alla Karpova, Janelia), a recently developed retrogradely transported AAV (Tervo et al., 2016). Subsequently, ketamine-anesthetized mice were head-fixed and laser stimulus grids of varying size and resolution were oriented either over the labeled corticospinal neurons in motor cortex or to cover as much of cortex as accessible. Cortical sites in the grid were sequentially stimulated by tcLSPS by steering the beam of a blue laser with a pair of scanning galvanometers (Thorlabs), using a pair of acousto-optical modulators for power control. Videos of the evoked movements were simultaneously captured with a pair of USB3 CMOS video cameras (Chameleon3, FLIR) running at 100 or 250 fps. Laser parameters and video capture were controlled using LabView. A simple, offline, ROI-based, 2D cross-correlation-based motion tracking algorithm written in MATLAB was used to measure movements of each paw from the recorded videos. The results demonstrate the utility and efficacy of tcLSPS-VMT for detecting and characterizing cortically evoked limb movements, and furthermore demonstrate that such movements can be evoked by selectively activating corticospinal neurons.

Disclosures: **L. Lambot:** None. **J.M. Barrett:** None. **X. Li:** None. **G.M. Shepherd:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.13/LL14

Topic: E.04. Voluntary Movements

Title: Oscillatory activity of supplementary motor areas reflects temporal integration of periodic events

Authors: *J. D. CADENA-VALENCIA, V. DE LAFUENTE
Inst. De Neurobiología, Querétaro, Mexico

Abstract: The perception of periodic events is essential to estimate incoming changes and to prepare and coordinate a timed motor command. To do so, the motor system must encode and compute temporal patterns and coordinate a motor plan according to the estimated periodicity of events. It is known that the supplementary motor area (SMA) is a structure that contains populations of cells that show tuning profiles ranging from specific motor actions to specific

time intervals in the millisecond scale. But it is not known the general population dynamics that is followed in order to give a coordinated output. The objective of the present project was to understand the general dynamics of SMA by studying its local field potentials (LFP) when monkeys have to perceive and anticipate visual periodic changes. We trained 2 rhesus monkeys in a task with 2 phases. First (Presentation phase) the monkeys have to attend a circular visual target that changes between two locations periodically over time. Then the target vanishes and the monkeys have to estimate the current position of the target as a function of the elapsed time (Estimation phase). A go cue presented at pseudorandom times instructed the monkeys to respond with the hand in which location the target would be at that time; the monkeys could not make any movements with the hand nor the eyes until the cue was presented. While monkeys performed the task, LFP was recorded in the SMA. To observe oscillatory patterns in the LFP we performed time frequency maps using multitaper techniques. In the estimation phase we found that even in the absence of a visual stimulus, the Gamma band´s amplitude (30-40 Hz.) changed accordingly to the previously presented stimuli. Studying Gamma power on single trial basis we observed Gamma bursting activity in the times when the target would change from one location to the next. Also we detected a linear relationship between the elapsed time and the average amplitude of the observed Gamma bursts. The observed patterns of LFP from SMA suggest that this structure is important to integrate temporal patterns even in the absence of a motor response. Also at population level, Gamma band activity may reflect the integration of different populations of cells in order to prepare a timed motor command.

Disclosures: J.D. Cadena-Valencia: None. **V. de Lafuente:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.14/LL15

Topic: E.04. Voluntary Movements

Support: Google Unrestricted Gift

Title: Motor cortical encoding of arm impedance during the coordinated control of both force and movement

Authors: *S. D. KENNEDY[1], A. B. SCHWARTZ[2]
[1]Bioengineering, Univ. of Pittsburgh, Pittsburgh, PA; [2]Dept Neurobiol, Univ. of Pittsburgh Dept. of Neurobio., Pittsburgh, PA

Abstract: The coordinated control of both force and movement is fundamental to object manipulation. However, the relation between force and movement can be unstable or unpredictable, making these manipulations difficult to perform. An impedance control

framework can simplify this problem. In this study, we tested the idea that arm impedance may be encoded in the neural activity of the motor cortex, with the implication that it is explicitly controlled by the motor system. We trained a monkey to pull on a handle that was locked in place until a specific force threshold was crossed. The threshold crossing triggered the handle to be unlocked so that it could be moved along a linear track. The monkey was required to stop the handle in one of four different targets. We found that arm impedance increased for targets that required shorter movements. For comparison, we repeated this procedure for four different force thresholds and found that arm impedance also increased for higher thresholds. This pattern was consistent with the framework of impedance control and led to our specific hypothesis: if a neuron encodes information about arm impedance, then that information should be consistent across thresholds and targets. Indeed, we found that 20 of 101 neurons had firing rates that were correlated with both target and threshold. In addition, 18 of those neurons had correlations consistent with arm impedance encoding. Specifically, the firing rates of 8 neurons were negatively correlated with target and positively correlated with threshold, i.e. positively correlated with arm impedance. The remaining 10 neurons had the opposite correlation. These results demonstrate that the simultaneous encoding of both force and movement in the firing rates of motor cortical neurons correspond to the variation of arm impedance during object manipulation and suggests that impedance control could be implemented by the motor system. Consideration of arm impedance can provide a concrete description of the coordinated control of force and movement.

Disclosures: **S.D. Kennedy:** None. **A.B. Schwartz:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.15/LL16

Topic: E.04. Voluntary Movements

Support: NSF CAREER IOS1553252

NSF Brain Initiative Grant (BCS1533672)

NIH R01-HD071686

NIGMS Training Grant T32GM081760

Bradford and Diane Smith Graduate Fellowship

Title: Comparing M1 neural reorganization during contralateral and ipsilateral visuomotor rotation learning

Authors: *L. BAHUREKSA[1,2], W. JOINER[3], S. CHASE[1,2]
[1]Dept. of Biomed. Engin., [2]Ctr. for the Neural Basis of Cognition, Carnegie Mellon Univ., Pittsburgh, PA; [3]Dept. of Bioengineering, George Mason Univ., Fairfax, VA

Abstract: It is well known that neurons in primary motor cortex (M1) exhibit tuning to kinematics of the contralateral arm. Further, several studies have demonstrated changes in this neural tuning during adaptation of contralateral arm movements. It has also been shown that M1 neurons often exhibit tuning to kinematics of the ipsilateral arm, and it has been hypothesized that this ipsilateral representation may contribute to generalization of learning to the untrained limb (e.g., Kawashima et al, *Brain Res*, 1994; Anguera et al., *Brain Res,* 2007). However, it has yet to be demonstrated whether ipsilaterally-tuned neurons exhibit changes in neural tuning during adaptation of movements of the ipsilateral arm. Here we compared the changes in direction tuning of M1 neurons during learning with the contralateral and ipsilateral arm. We trained a rhesus macaque to perform five sets of 8-target center-out reaching movements in a virtual reality environment. The subject first performed center-out reaching movements using the contralateral and ipsilateral hands (separate sets, 160 trials per hand). The subject then performed a set of 160 trials to these 8 targets with either the ipsilateral or contralateral arm during application of a visuomotor rotation (VMR; +/-40°). Following this perturbation session, the subject again completed center-out reaches using the contralateral and ipsilateral arms. Cosine tuning curves were fit to the data from each session of the experiment, and tuning changes were quantified for baseline firing rate, modulation depth (amplitude of the tuning curve) and preferred direction (PD; direction of maximal firing of the M1 neuron). We found robust changes in PD during learning of the VMR, counter to the direction of perturbation (-29.49 +/- 13.6º) (SD), with smaller, less consistent changes in modulation depth (0.29 +/- 5.29) (SD). These learning-related changes in neural tuning were of comparable magnitude during learning with either the contralateral or ipsilateral arm. These data imply that reorganization of M1 neurons is not preferential to contralateral learning, which may provide a substrate for bimanual transfer of learning.

Disclosures: L. Bahureksa: None. **W. Joiner:** None. **S. Chase:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.16/LL17

Topic: E.04. Voluntary Movements

Title: Demarcation of subjective value from arousal during action observation in F5 neurons

Authors: *S. SPADACENTA[1], J. K. POMPER[1], F. BUNJES[1], M. A. GIESE[2], P. THIER[1]
[1]Hertie Inst. For Clin. Brain Res., Tuebingen, Germany; [2]Hertie Inst. For Clin. Brain Res. / CIN, Tuebingen, Germany

Abstract: It has been suggested that mirror neurons in premotor area F5 are involved in the valuation of grasping actions performed by others. It is, however, unclear if the observed modulation attributed to subjective value may not be better explained by differences in arousal. We therefore recorded a total of 190 mirror neurons from the ventral premotor cortex of two rhesus macaques tested in a paradigm that dissociates subjective value from arousal. This paradigm required the animals to observe a video of a human hand grasping an object followed by four possible endpoints. These endpoints were randomly chosen per trial. Knowledge of the expected endpoint was provided by a cue, presented at the beginning of the trial, before the onset of the video. The four endpoints were: delivery of a large reward (1.5 ml water), a small reward (0.5 ml), a small punishment (no reward but 0.5 sec of eccentric fixation) or a large punishment (no reward but 3-5 sec of eccentric fixation). We expected that the more salient cues (large reward, large punishment) would cause more arousal than the less salient ones, allowing the separation of arousal and value. Based on the monkeys´ choice behavior and heart rate changes, we could establish that this expectation was met. We found that only a few F5 mirror neurons were classifiable as either reflecting value or arousal and that this reflection was often unstable over time. In contrast, a clear signature of value, stable over time, became apparent when ignoring the diversity of responses of single neurons by subjecting the whole population to a dimensionality reduction analysis based on the multidimensional scaling approach. Interestingly, this reflection of value was also found in F5 neurons not classified as mirror neurons, characterized by a lack of responses to observed actions. This congruency suggests a role of F5 neurons in the valuation of environments in general, rather than a role confined to action observation.

Disclosures: S. Spadacenta: None. **J.K. Pomper:** None. **F. Bunjes:** None. **M.A. Giese:** None. **P. Thier:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.17/LL18

Topic: E.04. Voluntary Movements

Support: NIH DP2 HD087952

Title: Head-restraint does not change the cortical dependence of a sensorimotor task in mice

Authors: T. BOLLU[1], N. PRASAD[2], *J. H. GOLDBERG[2]
[1]Neurobio. and Behavior, [2]Dept. of Neurobio. and Behavior, Cornell Univ., Ithaca, NY

Abstract: Cortical control of behavior differs across species and behavioral paradigms [1]. In head-fixed animals, inactivation of motor cortex dramatically impairs reach and lick behaviors [2; 3; 4; 5], but freely-moving animals with permanent cortical lesions [6; 7] can execute seemingly comparable tasks spontaneously with little to no impairment. To our knowledge cortical function has not been tested in the same animals performing the same task across head-fixed and free experimental preparations. To test if restraint can affect experimental results, we trained head-fixed and head-free mice in an identical sensorimotor lick task and optogenetically silenced different motor cortical areas during behavior. Performance similarly depended on tongue but not forelimb motor cortex in both experimental conditions, demonstrating that restraint does not alter cortical control of a simple sensorimotor task.
References:
[1] R.N. Lemon, Descending pathways in motor control. Annu Rev Neurosci 31 (2008) 195-218.
[2] J.Z. Guo, A.R. Graves, W.W. Guo, J. Zheng, A. Lee, J. Rodriguez-Gonzalez, N. Li, J.J. Macklin, J.W. Phillips, B.D. Mensh, K. Branson, and A.W. Hantman, Cortex commands the performance of skilled movement. Elife 4 (2015) e10774.
[3] Z.V. Guo, N. Li, D. Huber, E. Ophir, D. Gutnisky, J.T. Ting, G. Feng, and K. Svoboda, Flow of cortical activity underlying a tactile decision in mice. Neuron 81 (2014) 179-94.
[4] M.J. Goard, G.N. Pho, J. Woodson, and M. Sur, Distinct roles of visual, parietal, and frontal motor cortices in memory-guided sensorimotor decisions. Elife 5 (2016).
[5] T. Komiyama, T.R. Sato, D.H. O'Connor, Y.X. Zhang, D. Huber, B.M. Hooks, M. Gabitto, and K. Svoboda, Learning-related fine-scale specificity imaged in motor cortex circuits of behaving mice. Nature 464 (2010) 1182-6.
[6] R. Kawai, T. Markman, R. Poddar, R. Ko, A.L. Fantana, A.K. Dhawale, A.R. Kampff, and B.P. Olveczky, Motor cortex is required for learning but not for executing a motor skill. Neuron 86 (2015) 800-12.
[7] K. Lashley, Studies of cerebral function in learning: V. The retention of motor habits after destruction of the so-called motor areas in primates. Archives of Neurology & Psychiatry 12 (1924) 249-276.

Disclosures: T. Bollu: None. **N. Prasad:** None. **J.H. Goldberg:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.18/LL19

Topic: E.04. Voluntary Movements

Title: Representation of egocentric space in the frontal orienting field

Authors: *H. LI[1], H. H. YIN[2]
[1]Duke Univ., durham, NC; [2]Dept. of Psychology and Neurosci., Duke Univ., Durham, NC

Abstract: The frontal orienting field(FOF) has been known to modulate orienting behavior in rodents. Lesion, stimulation and anatomical experiments suggested that the FOF is critical for orienting in navigation and decision making. Specifically, it was found that unilateral activations of FOF lead to orienting bias towards the contralateral side. How FOF output directly shapes the trajectories of orienting movements, however, is not well understood. Using optogenetics and 3D motion capture techniques, we were able to quantify the relationship between FOF neural activity and the trajectory of orienting behavior. We found that 1) stimulation to the FOF led to various head movements that resemble natural orienting movements in rodents; 2) stimulations of a specific location in the FOF caused orienting movements towards a specific location relative to the body, suggesting that the FOF contains a egocentric spatial map of destinations of orienting movements; 3) surprisingly, unilateral stimulation of FOF in either hemisphere was similar to bilateral stimulation, both resulting in orienting to a specific egocentric location. This result indicates that FOF in each hemisphere possibly contains symmetrical representations of bilateral spatial locations.

Disclosures: H. Li: None. **H.H. Yin:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.19/LL20

Topic: E.04. Voluntary Movements

Support: NIH RO1DE023816

Title: Encoding properties of neurons in the orofacial sensorimotor cortex during bite force generation at varying gapes

Authors: *F. I. ARCE-MCSHANE[1], K. TAKAHASHI[1], B. J. SESSLE[2], N. HATSOPOULOS[1], C. ROSS[1]
[1]Dept. of Organismal Biol. and Anat., Univ. of Chicago, Chicago, IL; [2]Univ. Toronto, Toronto, ON, Canada

Abstract: Human mastication relies on the precise control of the generation of bite force at varying degrees of mouth opening, i.e. gapes. The role of orofacial sensorimotor cortex in the coordination of gape and bite force is unknown. Here we examined the spiking activity recorded with microelectrode arrays implanted chronically in the primary motor (MIo), primary

somatosensory (SIo), and cortical masticatory (CMA) areas of the orofacial sensorimotor cortex in monkeys (*Macaca mulatta*) trained to generate one of three bite force levels at one of three gapes per trial. We used generalized linear models to predict the time-varying spiking activity of each neuron as a function of bite force, gape, and spike histories of neurons recorded from the same cortical area. To assess model performance, we computed the area under the receiver operating characteristic curve (AUROC) obtained from cross-validated test trials and compared the values obtained from the full model that has bite force, gape, and spike histories of neurons as covariates against the performance of reduced models that have the bite force or gape excluded. We found that both bite force and gape were predictive of the spiking activity of the population of neurons in MIo, SIo, and CMA as AUROC values of full and reduced models were significantly above chance level (Wilcoxon Signed Rank Test, $p<0.00001$). In MIo, model performance was significantly degraded when the bite force was excluded from the predictors (Wilcoxon Signed Rank Test, $p<0.01$) and when both bite force and gape were excluded (Wilcoxon Signed Rank Test, $p<0.01$). In SIo and CMA, the AUROC values in the reduced models were not significantly lower than those in the full model (Wilcoxon Signed Rank Test, $p>0.05$). Overall, the results suggest that while single-unit and population responses in all three areas play a role in the coordination of bite force and gape, MIo may perform computations that are different from those performed by SIo and CMA.

Disclosures: F.I. Arce-Mcshane: None. **K. Takahashi:** None. **B.J. Sessle:** None. **N. Hatsopoulos:** None. **C. Ross:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.20/LL21

Topic: E.04. Voluntary Movements

Support: Grant-in-Aid for Scientific Research (A)

Grant-in-Aid for Scientific Research for Innovative Area

JST PREST

Title: Presynaptic inhibition of muscle afferent input to spinal cord in awake, behaving monkeys

Authors: *S. TOMATSU[1], G. KIM[1], S. KUBOTA[1,2], K. SEKI[1,3]
[1]Natl. Inst. of Neuroscience, NCNP, Tokyo, Japan; [2]JSPS, Tokyo, Japan; [3]JST, PREST, Tokyo, Japan

Abstract: In the previous studies (Seki et. al. 2003 *Nat Neurosci* and 2009 *J Neurophysiol*), we have reported the modulation of the level of presynaptic inhibition on the input from cutaneous

afferent in monkey spinal cord during voluntary movement. In these report, we have developed the method to estimate the size of primary afferent depolarization by evoking antidromic volleys in the cutaneous nerve by applying the microstimulation to their intraspinal terminals (Wall's excitability testing). In the present study, we applied this technique to evaluate the modulation of presynaptic inhibition on a muscle afferent from wrist extensors in monkeys (n = 2) performing wrist flexion-extension task. After finishing the behavioral training, we implanted an oval chamber to the lower cervical vertebrae (C4-T1) for intraspinal microstimulation, a custom-made nerve cuff electrode to the deep radial nerve (DR) innervating wrist extensor muscles, and electromyographic electrodes to wrist and finger muscles. Using a tungsten microelectrode, we applied intraspinal microstimulation (10 Hz, 1-50 μA) to the ventral horn while monkeys are performing wrist movement to activate the terminal of DR afferents. In total, 49 volleys were recorded in the DR nerve of two monkeys (n = 38 and 11, respectively). Onset latency of each volley was distributed from 2.23 ms to 4.03 ms (mean 2.92 ± 0.47 ms and 3.23 ± 0.20 ms, respectively) suggesting a majority of volley were induced in the faster-conducting muscle afferents. In both monkeys, we found a significant reduction in the size of antidromic volleys in the DR nerve during wrist movements. In the first monkey, for example, task related reduction in their amplitude was found in ten among 28 putative antidromic volleys (35.7 %), and the reduction is found predominantly during isometric wrist extension ($p < 0.05$). In contrast, the facilitation in size (n = 6) of volley was not frequently observed in any specific epoch of the task. In the second monkey, the size of antidromic volleys was also suppressed when the monkey performed dynamic and isometric wrist extension ($p < 0.05$) in 11 putative antidromic volleys, so far. These results suggested that the level of presynaptic inhibition on a muscle nerve is suppressed during agonistic torque. Such disinhibition might allow the spinal cord to use the information from muscle afferent in an efficient way for the control of ongoing movement. Based on our previous finding on the facilitation of presynaptic inhibition on the cutaneous afferent during active movement (Seki et al. 2003, 2009), we can now conclude that task-relevant peripheral inputs are highlighted by the presynaptic inhibition in a highly selective way.

Disclosures: **S. Tomatsu:** None. **G. Kim:** None. **S. Kubota:** None. **K. Seki:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.21/LL22

Topic: E.04. Voluntary Movements

Support: NRF-2016M3C7A1904986

Title: Decoding neuronal firing patterns in the anterior lateral motor cortex into sensory cue information

Authors: *J.-W. SOHN[1], S. CHAE[2], S.-P. KIM[3]
[1]Daegu-Gyeongbuk Med. Innovation Fndn., Daegu, Korea, Republic of; [2]Design and Human Engin., Ulsan Natl. Inst. of Sci. and Technol., Ulsan / Ulsan Metropolitan City, Korea, Republic of; [3]Ulsan Natl. Inst. of Sci. and Technol., Ulsan, Korea, Republic of

Abstract: Neurons in the anterior lateral region of the motor cortex (ALM) of rodents have been known to modulate their spiking activity according to conditioned sensory input from the whisker. However, the role of ALM in the flow of information from decision to action is not well known. The present study aims to address this by decoding neuronal firing patterns in the ALM to extract the information from sensory cues. For this, we analyzed single-unit activities recorded in the left ALM of rodents which were available from the CRCNS depository for public use. For the behavioral task, three mice performed the object discriminant task with a tactile sensory cue. In each task trial, a pole touched (1.3 s) a posterior whisker for indication of upcoming water reward from the right frontal side of the mice, and an anterior whisker from left frontal side (stimulus-on period). After the pole out, the mice were forced to wait for 1.3 s (delay period). Then, the mice licked to right or left based on memory (peri-movement period). The behavioral outcomes were divided into four types: hit right, hit left, error right and error left. The neuronal data were recorded from 32 channel micro electrode array while the mice performed the task. We decoded the sensory cue information (i.e. posterior vs. anterior) from the firing rates of the ensemble of 10 to 20 neurons using the linear discriminant analysis (LDA). We applied LDA for each of three periods with groups of neurons showing selectivity for sensory stimuli ($p<0.01$) based on mutual information. In addition, we tested the performance of decoder trained using either the hit trial data or error trial data and cross-validated to prevent overfitting. During the stimulus on period, the predicted accuracy of sensory cue of decoder trained with hit trials (error trials) was 49.60% (53.08%). In contrast, the performance of decoder was 78.72% (78.33 %) and 86.28% (82.05%) respectively during delay period and peri-movement period. The decoding analysis showed that we could decode neuronal firing patterns into sensory cue information significantly higher than the chance level (50%) during delay period and peri-movement period but not during stimulus on period. This tendency was preserved regardless of training data. This result implies that that the role of ALM may be related to retention of working memory and movement execution rather than decision making for sensory cue.

Disclosures: J. Sohn: None. **S. Chae:** None. **S. Kim:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.22/LL23

Topic: E.04. Voluntary Movements

Support: Howard Hughes Medical Institute

Title: Clustering of dendritic activity during decision making

Authors: A. M. KERLIN, *B. MOHAR, D. A. FLICKINGER, B. J. MACLENNAN, C. DAVIS, N. JI, N. SPRUSTON, K. SVOBODA
HHMI Janelia Res. Campus, Ashburn, VA

Abstract: Neighboring neurons in motor cortex exhibit diverse selectivity during sensation, movement preparation, and movement execution. Neuronal selectivity could emerge from diverse mechanisms, including selective connectivity and nonlinear interactions of synaptic inputs in dendrites. We studied dendritic integration in the anterior motor cortex of mice performing a tactile discrimination task with a delayed response (Guo and Li et al., 2014). We constructed a two-photon microscope that allows rapid (~15 Hz) imaging of up to 300 µm of contiguous dendrite while resolving calcium transients in individual dendritic spines. Two galvanometers and a remote focusing mirror (Botcherby et al., 2008) steer 16 kHz lines (24 µm extent) produced by a resonant mirror arbitrarily in three dimensions. Pyramidal neurons were labeled sparsely with GCaMP6f in transgenic mice. We imaged spine and dendritic calcium transients, as well as somatic calcium transients associated with action potentials. We developed methods to computationally remove the influence of backpropagating action potentials (bAPs), which allowed us to quantify the selectivity of spines and dendritic segments during sensation, movement preparation, and movement execution. Nearby spines and dendritic segments share similar selectivity (length constant of signal correlation, ~30 µm). This clustering was more often seen in distal than in proximal dendrites. Using a measure of local autocorrelation, we also found that this reflects distinct “hotspot” locations on the dendrite where nearby dendrite and spines are co-active in time. Hotspot selectivity was correlated with the behavioral selectivity of somatic spikes, suggesting that these locations may have privileged influence over the output of the cell.

Disclosures: A.M. Kerlin: None. **B. Mohar:** None. **D.A. Flickinger:** None. **B.J. MacLennan:** None. **C. Davis:** None. **N. Ji:** None. **N. Spruston:** None. **K. Svoboda:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.23/LL24

Topic: E.04. Voluntary Movements

Support: NIH Grant NS078127

The Sloan Foundation

The Klingenstein Foundation

The Simons Foundation

The Center for Sensorimotor Neural Engineering

The McGovern Institute

Title: Predictive coding of temporal events through regulation of cortical dynamics

Authors: *S. W. EGGER[1,2], C.-J. CHANG[1,2], M. JAZAYERI[1,2]
[1]McGovern Inst. for Brain Res., [2]Brain and Cognitive Sci., MIT, Cambridge, MA

Abstract: Natural sensorimotor behavior relies on flexible and rapid integration of incoming sensory information with ongoing motor plans. To gain insight into the neural mechanisms of sensorimotor integration, we recorded from the dorsomedial frontal cortex (dMFC) of monkeys performing a novel timing task. Animals had to integrate information from the first three beats of a rhythm (S1, S2, and S3) in order to initiate a saccade at the expected time of the missing fourth beat (Go). Analysis of behavior indicated that animals integrated information from the S1-S2 and S2-S3 epochs to optimize their response, consistent with a Bayesian estimation model. We considered two algorithms the brain might use to perform this integration. The first was a sequential estimation algorithm in which the estimate is updated after each beat, and the second was batch estimation algorithm in which the estimate is derived in a single step after S3. Although dMFC responses were strongly modulated throughout the trial, computational principles of Bayesian estimation were not discernable at the level of single neurons. However, hallmarks of sequential updating algorithm emerged from an analysis of dynamics at the population level. Across task epochs, population trajectories resided in different regions of the state space, and within each epoch, they were systematically organized with respect to the inter-beat-interval. This organization exhibited two characteristics: 1) the speed with which the state evolved along the trajectory predicted the time of the upcoming beat, and 2) the neural state at the time of each beat encoded a prediction error signal. Importantly, error signals at the end of each epoch predicted the speed of population dynamics during the ensuing epoch. We captured these features in a cascade of two-neuron models, each capable of performing a canonical operation of converting an error signal to predictive dynamics by adjusting the speed. These results suggest that cortical circuits integrate sequentially presented information by implementing an error-based sequential updating mechanism, akin to predictive coding. More generally, our work highlights the importance of inferring algorithms of sensorimotor and cognitive computations at the level of population dynamics.

Disclosures: **S.W. Egger:** None. **C. Chang:** None. **M. Jazayeri:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.24/LL25

Topic: E.04. Voluntary Movements

Support: NIH Grant U01EB017695

NIH Grant R01EB022903

NIH Grant R01MH086638

NYS Grant DOH01-C32250GG-3450000

Title: Long-range inputs and H-current regulate different modes of operation in a multiscale model of mouse M1 microcircuits

Authors: *S. DURA-BERNAL[1], S. A. NEYMOTIN[2], B. A. SUTER[3], G. M. SHEPHERD[4], W. W. LYTTON[5]
[1]Physiol. and Pharmacol., State Univ. of New York Downstate Med. Ctr., Brooklyn, NY; [2]Dept. Neurosci., Brown Univ., Providence, RI; [3]Physiol., [4]Dept Physiol., Northwestern Univ., Chicago, IL; [5]SUNY Downstate, Brooklyn, NY

Abstract: We hypothesize the primary motor cortex (M1) may operate in different modes, or along a continuum of modes, characterized by the degree of activation of intratelencephalic (IT), corticothalamic (CT) and pyramidal tract (PT) neurons. The "IT/CT-predominant" mode would involve highly active IT and CT neurons but low PT activity; whereas the "PT-predominant" would exhibit the opposite pattern. In the "IT/CT" mode, a high level of H-current in PT neurons would modulate synaptic integration of inputs and reduce PT output activity. Downregulation of the H-current would lead to increased PT activity in the "PT" mode. The particular subset of M1 neurons targeted by different long-range inputs could also play a role in regulating the different modes: thalamic posterior nucleus (PO) and somatosensory cortical (S1 and S2) inputs would favor the "IT/CT" mode, whereas thalamic ventro-lateral (VL) and motor cortical (contralateral M1 and M2) would promote the "PT" mode.To study this hypothesis we developed a multiscale data-driven model of mouse M1 microcircuits, with over 10,000 cells distributed in a cylindrical volume of diameter 300 μm and cortical depth 1350 μm. L5 IT and PT neuron morphologies with 700+ compartments reproduced cell 3D reconstructions, and their ionic channel distributions were optimized within experimental constraints to reproduce in vitro recordings. The network includes over 30 million synaptic connections that depend on pre- and post-synaptic cell class and cortical depth. Data was based on optogenetic circuit mapping studies which determined that connection strengths vary within layer as a function of the neuron's cortical

depth. The synaptic input distribution across cell dendritic trees -- likely to subserve important neural coding functions -- was also mapped using optogenetic methods and incorporated into the model. The network was driven by the main long-range inputs to M1: thalamus PO and VL, S1, S2, contralateral M1, M2, and orbital cortex (OC). We studied the effect on M1 of increased activity in each of these regions, and of different levels of H-current in PT neurons. Microcircuit dynamics and information flow were quantified using firing rates, oscillations, and information transfer measures (Granger causality and normalized transfer entropy). Preliminary results support the different modes of operation hypothesis; further exploration will help characterize the underlying mechanisms and determine interactions between the factors involved.

Disclosures: S. Dura-Bernal: None. **S.A. Neymotin:** None. **B.A. Suter:** None. **G.M. Shepherd:** None. **W.W. Lytton:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.25/LL26

Topic: E.04. Voluntary Movements

Support: NIH grant FNS070366A

NIH Grant P30 NS076405

Title: Primary motor cortex neurons which produce post-spike suppression provide an active descending command to turn off muscle activity

Authors: *D. M. GRIFFIN, D. S. HOFFMAN, P. L. STRICK
Systems Neurosci. Inst., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Step-tracking movements are characterized by distinct spatiotemporal patterns of activity in agonist and antagonist muscles. Before the agonist burst of activity and the subsequent generation of movement occurs, there is a distinct phasic decrease in antagonist muscle activity. This premovement antagonist inhibition precedes the agonist burst by 50 milliseconds and has been termed the Hufschmidt phenomenon (Hufschmidt and Hufschmidt, 1954). It is generally accepted that the agonist and antagonist bursts of muscle activity are centrally generated. Here we provide evidence that the Hufschmidt phenomenon is also centrally generated. To investigate the origin of premovement antagonist inhibition, we examined primary motor cortex (M1) neurons that produced post-spike suppression effects in averages of electromyographic (EMG) activity. We trained a rhesus monkey to perform a center-out step tracking wrist task. Then, we collected neural activity from M1 and EMG activity from 12 task related wrist and digit muscles. We used spike-triggered averaging of EMG activity to identify 20 M1 neurons that produced

post-spike suppression in at least one recorded muscle. Eight of these neurons (40%) showed a phasic increase in activity when their target muscle functioned as an antagonist. These eight M1 neurons displayed phasic bursts of activity that occurred at the same time or slightly before the phasic decrease in the activity of their target muscle. Thus, the phasic activity of M1 neurons which produce disynaptic post-spike suppression of muscle activity clearly contribute to and may even generate the Hufschmidt phenomenon. Our results provide further evidence that M1 neurons are functionally tuned to sculpt distinct spatiotemporal patterns of muscle activity.

Disclosures: D.M. Griffin: None. **D.S. Hoffman:** None. **P.L. Strick:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.26/LL27

Topic: E.04. Voluntary Movements

Support: HHMI

Helen Hay Whitney Foundation

Title: Discrete attractor dynamics underlies selective ramping activity in frontal cortex

Authors: *H. INAGAKI, L. FONTOLAN, S. ROMANI, K. SVOBODA
JFRC, Ashburn, VA

Abstract: Short-term memory links past experience and future actions. Short-term memory is represented by changes in spike rates that are maintained internally. Neuronal time constants are typically on the order of milliseconds. Persistent and ramping activity related to short-term memory can last many seconds. The underlying mechanisms could be related to cellular bistability, chain-like sequential excitation in the circuit, or different types of circuits with positive feedback. We studied the mechanisms of short-term memory in a delayed response task. During the delay epoch, neurons in the mouse anterior lateral motor cortex (ALM) show monotonic and low-dimensional persistent activity that instructs future actions. To distinguish between different neural circuit mechanisms underlying selective persistent activity we performed whole-cell recordings and high-density silicon probe recordings in ALM. We found no evidence for cellular intrinsic bi-stability. Instead, both membrane potentials and population spiking dynamics funneled toward discrete endpoints during the delay epoch. The discrete endpoints were robust to shifts in population dynamics caused by optogenetic perturbation. Perturbation occasionally resulted in switching of the dynamics to the other endpoints, followed by incorrect actions. Our results support the idea that discrete attractors underlie short-term memory in frontal cortex.

Disclosures: **H. Inagaki:** None. **L. Fontolan:** None. **S. Romani:** None. **K. Svoboda:** None.

Poster

407. Cortical Planning and Execution: Animal Neurophysiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 407.27/LL28

Topic: E.04. Voluntary Movements

Title: Models for short-term memory in a motor preparation task

Authors: ***L. FONTOLAN**, H. K. INAGAKI, K. SVOBODA, S. ROMANI
Janelia Res. Campus, Howard Hughes Med. Inst., Ashburn, VA

Abstract: Short-term memory (STM) is the ability to temporarily hold stimulus-related information in order to carry out cognitive tasks. At the core of STM maintenance is the question of how to sustain a representation after the stimulus is removed. Persistent, stimulus-dependent neuronal activity has been observed in numerous brain areas during tasks requiring the temporary maintenance of information, either related to external stimuli or motor planning. Neural recordings performed during STM tasks typically reveal the presence of populations of neurons that display selective sustained activity during the delay epoch. Given that these persistent states can last seconds, i.e. orders of magnitude longer than the typical neuronal time constants, proposed mechanisms to explain the experimental evidence either rely on single-cell multistability or require circuit-level interactions. In this work, we tested the ability of these models to account for electrophysiological data in premotor cortex during a binary delayed-response task in rodents. Analysis of these data excluded single-cell multistability and neuronal chains, where information is passed along each node in a chain of connected neurons. Two major classes of STM models could still potentially account for the observed activity:
i) Positive or negative-derivative feedback transiently maintaining a memory trace by slowing down the decay towards a low-firing stable state.
ii) Strong positive feedback giving rise to a high-firing attractor state, where memory is maintained by balancing decay through reverberating excitatory loops.
Additionally, the firing patterns described above could generate trajectories of various dimensionality (e.g. point, line, etc) in the space of network population activity. To discriminate between these models, we generated predictions for network behavior during non-selective optogenetic manipulations, similar to those carried out experimentally. Models in (i) could be ruled out, since their dynamics would reflect the integration of the optogenetic manipulation, which was not observed in the data. Instead, models in (ii), implemented in our case as a bistable attractor network, could accurately reproduce the observed neuronal dynamics in both control and perturbed conditions. Our combined computational and experimental approach reveals the presence of a stable fixed point encoding the task-relevant information during the preparation of

motor actions. The formation of discrete attractors may be a general mechanism underlying STM in cortical circuits.

Disclosures: L. Fontolan: None. **H.K. Inagaki:** None. **K. Svoboda:** None. **S. Romani:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.01/LL29

Topic: E.04. Voluntary Movements

Support: NIH Grant R21DC012502

Title: Modulation of event-related potentials associated with orofacial skin stretch during speech production

Authors: *T. ITO[1,2], H. OHASHI[3], E. MONTAS[2], V. L. GRACCO[2,4]
[1]Gipsa Lab, CNRS, St Martin D Heres Cedex, France; [2]Haskins Labs., New Haven, CT; [3]Haskins Labs. Inc, New Haven, CT; [4]McGill Univ., Montreal, QC, Canada

Abstract: Modulation of sensory event-related potentials occurs before or during voluntary movement. For speech, the amplitude of auditory cortical potentials is reduced during speech production, the so-called speech induced suppression. However, it is unknown whether somatosensory processing is also suppressed during speech production. The current study addressed this question by examining whether somatosensory event-related potentials (ERPs) associated with facial skin deformation are changed during speech production tasks. We investigated changes in the magnitude of somatosensory ERPs during speech posture with and without voicing, and also with changes associated with different speech utterances. ERPs from 64 scalp sites in response to somatosensory stimulation associated with facial skin stretch were recorded. Participants engaged in: 1) a vowel production task, and, 2) a non-speech task in which participants maintain the same posture without voicing. Three vowels (/a/, /i/, and /u/) were used in these two production tasks. Somatosensory ERPs in the six task conditions (3 vowels x 2 production tasks) were compared with the ERPs from control (resting) condition. We applied global field power (GFP) measures to evaluate the amount of activity of each time point for the ERPs on the cortical surface. GFP showed two peaks consistently around 160 ms and 320 ms after somatosensory onset in all conditions. The first peak amplitude was significantly different for the three vowel tasks, but there were no significant changes in the second peak. The first peak in the vowel /u/ was reliably smaller than those in the other two vowels. This was found consistently in both vowel production and posture tasks. Cluster analysis of the temporal pattern of the GFP showed a significant reduction of somatosensory GFP in the /u/ condition relative to the control condition, while the other two vowel conditions were not reliably different.

Displacement of facial skin stretch was not different across the three vowel tasks, indicating that the changes of ERP responses were not attributed to a magnitude of the facial skin deformation. Taken together, the sensitivity of somatosensory ERP associated with facial skin deformation changes according to the specific vowel being produced. More importantly, there is evidence for a short latency (around 160 ms) suppression of somatosensory input during speech. It appears that sensory suppression mechanisms are reflected in both sensory modalities (auditory and somatosensory) associated with speech production and the somatosensory system may be modulated differently relative to phonetic identity.

Disclosures: **T. Ito:** None. **H. Ohashi:** None. **E. Montas:** None. **V.L. Gracco:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.02/LL30

Topic: F.05. Neuroimmunology

Support: University of Nebraska at Omaha Office of Research and Creative Activity

Title: Lingual nerve transection induces neutrophil response in the anterior tongue of rats

Authors: *J. D. OMELIAN[1], S. I. SOLLARS[2]
[1]Psychology, Univ. of Nebraska at Omaha Dept. of Psychology, Omaha, NE; [2]Univ. of Nebraska At Omaha, Omaha, NE

Abstract: The anterior tongue is maintained by bilateral gustatory (chorda tympani) and somatosensory (lingual) nerves which innervate tissue in close physical proximity. These nerves appear to have a cooperative, multimodal relationship, in which loss of one nerve results in corresponding changes to the tissue (e.g., taste bud or fungiform papillae) associated with the other. Regardless of which nerve is transected, changes in morphology occur primarily on the denervated side of the tongue, with the contralateral intact side serving as control tissue. One possible mechanism for the observed multimodality of nerve transection effects may be in the inflammatory response following nerve injury. To investigate this possibility, we quantified the innate immune response in the anterior tongue following transection of the lingual nerve (LX), leaving the chorda tympani intact. Adult, female Sprague-Dawley rats underwent unilateral LX at 65 days of age and were allowed to recover for 12 or 24 hours. Following sacrifice, tongue tissue was collected, frozen, and sectioned. Immunohistochemistry was used to visualize myeloperoxidase positive neutrophils stained with DAB, and counts were taken from 10 μm sections within the first 1.5 mm of the anterior tongue. Neutrophil counts were compared between the intact and denervated sides of the tongue. Tissue from two age-matched nonsurgical animals was also processed, counted, and used for additional comparison. Preliminary results

suggest that average neutrophil counts do not differ between intact and cut sides of the tongue at 12 or 24 hours following unilateral LX in adult rats. Significantly more neutrophils were present on the denervated side of the tongue tissue than were present in non-surgical control tissue. Interestingly, the average number of neutrophils on the intact side of the tongue was also significantly higher than in non-surgical tongue tissue at 24 hours post-surgery, suggesting that LX may invoke an immune response which is not restricted to the denervated side of the tongue. This is in contrast with the changes in morphology reported following lingual or chorda tympani transection, which are typically restricted to the injured side of the tongue. Additional comparisons (e.g., animals that undergo sham surgery) will be needed to clarify the effect of LX surgery on neutrophil response in the anterior tongue.

Disclosures: J.D. Omelian: None. **S.I. Sollars:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.03/LL31

Topic: E.04. Voluntary Movements

Support: ANR Grant 16-CE37-0007-03

Title: Motor control of the tongue during speech: Predictions of an optimization policy under sensorimotor noise

Authors: *P. BARADUC, P. PERRIER
Dpt Speech and Cogntion, Gipsa-Lab, CNRS / U. Grenoble-Alpes UMR 5216, Saint-Martin d'Hères, France

Abstract: Speech production necessitates a high dexterity. Some sounds like /i/, /s/ or /l/ require every precise positioning of the tongue, while tasks constraints can vary widely (inertial perturbations in walking/running, vocal tract perturbations during eating...). Moreover, tongue movements are very rapid, on the order of some tenth of millisecond, while proprioceptive and auditory feedback latencies are longer. Last, sensory and motor signals have limited precision (they are 'noisy'). For all these reasons, it has been proposed that the central nervous system (CNS) rely on an optimal estimation system ('internal model') in order to adjust the trajectories in real time. Moreover, the CNS also seems to optimize some perceived cost, as suggested by the motor stereotypies observed in eye or arm movements. In particular, the hypothesis of the minimization of an internal measure of effort, or the minimization of the impact of motor noise on endpoint variance account for a large body of experimental observations. Here we test whether an effort optimization controller coupled to an optimal state estimator can account for the trajectories of the tongue when a subject produces the three vowels /i/, /a/ and /u/ from a

neutral (schwa) initial posture. In a first step, we ran 20,000 simulations of a finite element model of the tongue in order to describe the effect of combinations of muscle activations ramps across six muscles of the tongue (3 intrinsic, 3 extrinsic). We then applied model identification techniques to obtain a computationally tractable dynamical model of the tongue in the sagittal plane. Assuming a fixed jaw position and a standard geometry for the rest of the vocal tract, we obtained a simplified model of the speech production system. We could then derive the first three formants of the voice from the instantaneous tongue position through a harmonic analysis. With the sensorimotor plant thus defined, we applied standard numerical techniques inside a time loop to simulate the function of an optimal estimator/controller subjected to sensory and motor uncertainty, and generated tongue trajectories from the initial posture to the final endpoints defined either in postural space or in acoustic (F1-F2) space. These simulations allow exploring how optimal control hypotheses can explain the average trajectories and the impact of sensorimotor noise on their variability.

Disclosures: P. Baraduc: None. **P. Perrier:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.04/LL32

Topic: E.04. Voluntary Movements

Support: JSPS Grant 15H05880

Title: Entrainment of chewing rhythm by gait rhythm during treadmill walking in humans

Authors: H. MAEZAWA[1], S. KOGANEMARU[2], M. MATSUHASHI[3], M. FUNAHASHI[4], *T. MIMA[5]
[1]Hokkaido Univ., sapporo, Japan; [2]Hokkaido Univ., Sapporo, Japan; [3]Human Brain Res. Ctr., Kyoto Univ. Grad. Sch. of Med., Kyoto-Shi, Japan; [4]Hokkaido Univ, Grad Sch. Dent. Med., Sapporo, Japan; [5]Ritsumeikan Univ., Kyoto, Japan

Abstract: Autonomous rhythmic motor behaviors such as chewing and gait maintenance are critical for daily human life. These rhythmic motor behaviors are realized in different frequency bands for different body parts (i.e., those in the facial area for chewing and foot area for gait), suggesting that they are regulated by independent central mechanisms. However, it remains unclear whether the rhythmic processes of chewing and gait synchronize during concurrent execution in humans. To evaluate the entrainment of chewing rhythm by gait speed, we measured electromyography (EMG) signals from masticatory muscles and tibialis anterior (TA) muscles during chewing at a habitual rhythm while walking on a linear treadmill in 13 healthy volunteers. The up-and-down movement of the head was also measured using an accelerometer

on the back of neck. Each 6-min session included gait tasks performed at three speeds using treadmill (participant's self-selected gait speed: Gait-Auto, Gait-Auto × 1.3: Gait-High, Gait-Auto ÷ 1.3: Gait-Low). For control, we also measured EMG signals from masticatory muscles during chewing while stationary (Chew-Only) before the gait tasks. Chewing rhythm during walking was the same as that for head movement, occurring at twice the speed of the walking rhythm, in nine participants in the Gait-Auto condition, eight participants in the Gait-High condition, and eight participants in the Gait-Low condition. For these subjects, chewing rhythm in the Gait-Auto and Gait-High conditions differed significantly from that in the Chew-Only condition. However, no significant differences in chewing rhythm were observed between the Gait-Low and Chew-Only conditions. Significant differences of chewing rhythm were also observed between the Gait-Auto and Gait-High conditions, between the Gait-Auto and Gait-Low conditions, and between the Gait-High and Gait-Low conditions. Coherence between masticatory muscles and head movements was detected in all gait conditions (Gait-Auto, Gait-High, Gait-Low). Our findings therefore suggest that habitual chewing rhythm is entrained by gait rhythm, which may be mediated by the interaction between two central rhythm generators, or by head movement associated with walking via peripheral mechanisms.

Disclosures: H. Maezawa: None. **S. Koganemaru:** None. **M. Matsuhashi:** None. **M. Funahashi:** None. **T. Mima:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.05/LL33

Topic: E.04. Voluntary Movements

Support: NIH Grant R01 DC002852

Title: Somatosensory-based compensation to mechanical perturbations of the larynx during speech

Authors: *D. J. SMITH[1], A. F. SALAZAR-GOMEZ[2], C. E. STEPP[1], F. H. GUENTHER[1]
[2]Grad. Program for Neuroscience. Computat. Neurosci., [1]Boston Univ., Boston, MA

Abstract: When a brief, unexpected perturbation to a talker's auditory feedback gives the perception that voice fundamental frequency (f0) has decreased, he/she typically compensates by increasing f0 through auditory feedback control mechanisms. This compensatory response is only partial, typically less than 30% of the size of the perturbation. A brief, unexpected mechanical perturbation to the larynx (a somatosensory perturbation) causes a compensatory response in a manner similar to auditory perturbation. However the compensatory responses to somatosensory perturbations are greater than typically seen for auditory perturbations. Prior

studies of somatosensory perturbation of the larynx did not mask auditory feedback; thus it is unclear whether somatosensory feedback control mechanisms alone are responsible for this increased compensatory response. In the current study, we applied short (1-1.5 s) physical displacements of the larynx using a specially designed device while participants phonated the vowel /i/. To minimize the use of auditory feedback control mechanisms, participants received 90 dB speech-shaped masking noise over headphones. We found that displacing the larynx caused an initial decrease in f0 followed by a compensatory increase. When the larynx was released, f0 increased initially, then decreased toward the baseline value. These compensatory responses, mediated by somatosensory feedback control mechanisms in the absence of auditory feedback control, were much larger than typically seen in auditory perturbation studies, approaching 90% compensation within 1 second of the perturbation. There are at least two possible explanations for this discrepancy between compensatory responses to auditory versus somatosensory perturbations. First, the gain of the auditory feedback controller may be lower than the gain of the somatosensory feedback controller, possibly because longer delays in the auditory feedback system require a lower gain to maintain stability. Second, compensation to an auditory perturbation naturally leads to somatosensory feedback signals that do not match the speaker's somatosensory target, resulting in competition between the auditory and somatosensory feedback controllers. This competition is not present when somatosensory feedback is perturbed because the perturbation also perturbs auditory feedback in the same direction, allowing the speech motor control system to more completely compensate for the perturbation through somatosensory feedback control mechanisms.

Disclosures: D.J. Smith: None. **A.F. Salazar-Gomez:** None. **C.E. Stepp:** None. **F.H. Guenther:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.06/MM1

Topic: E.04. Voluntary Movements

Support: ERC 320708 (iCONNECT)

Title: Speech movement parameters reflected in sensorimotor cortex activity

Authors: *E. SALARI, Z. V. FREUDENBURG, M. J. VANSTEENSEL, N. F. RAMSEY
Neurol. & Neurosurg., Univ. Med. Ctr. Utrecht, Utrecht, Netherlands

Abstract: Although the sensorimotor cortex is known to be an important area for controlling movements, neuronal activity in this area is not always linearly related to motor output. When repeating a finger movement, for example, neural activity declines over repetitions, despite equal

performance (Hermes et al. 2012). It is unknown whether other movements, such as those involved in speech, show a non-linear relationship with underlying neural activity. Insight in this matter will contribute to our understanding of speech production, but is also relevant for the development of speech-based brain-computer-interfaces.
Here, we investigated brain activity during repeated speech movements with electrocorticography (ECoG) over the sensorimotor mouth area. Four epileptic patients who were implanted with ECoG electrodes for diagnostic purposes participated in our study. Two subjects agreed to have an extra high-density electrode grid for research purposes. Subjects were asked to repeat the Dutch /i/ sound at 3 different rates (5,4 or 3 repetitions in 3 seconds). A trial started with an indication of the speech rate, followed by speech cues (at the same rate, visually presented). Conditions were repeated 13 times, in random order.
Brain data was preprocessed (noisy electrodes and line-noise removed, common average re-referenced) and the high frequency band (HFB; 65-135Hz) power was extracted and temporally smoothed for all electrodes that displayed significant correlation to the task. The audio signal was used to extract information about speech movement parameters (sound intensity, repetition rate, tongue position and lip rounding), which were included as independent variables in the analyses (compensation for variations across trials). Subsequently, for every /i/ sound repetition, the peak height of brain activity was determined and averaged over trials, before and after compensation for speech movement variations.
We found that, without considering movement parameter variations, brain activity decreased across movement repetitions for almost all subjects and movement rates. This decrease disappeared when movement parameters were included in the analysis. This indicates that the variability in neural activity across repetitions is associated with differences in speech movement parameters.
The results suggest that the activity in the sensorimotor mouth area is closely linked to motor output. As opposed to earlier findings for finger movements, repetitions of speech movements did not clearly affect the amplitude of the HFB response. The apparent difference between finger and mouth movement encoding warrants further research.

Disclosures: E. Salari: None. **Z.V. Freudenburg:** None. **M.J. Vansteensel:** None. **N.F. Ramsey:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.07/MM2

Topic: E.04. Voluntary Movements

Support: NIH/NIDCD Grant R01DC014510

Title: Sensorimotor adaptation of speech with delays in the formant-shifted auditory feedback signal

Authors: T. MITSUYA[1], D. SHILLER[2], *L. MAX[1]
[1]Univ. Washington, Seattle, WA; [2]Ecole d'orthophonie et d'audiologie, Univ. of Montreal, Montreal, QC, Canada

Abstract: One potential problem for efficient motor learning arises when movement-related feedback is delayed. It is indeed well documented that perceiving the consequences of one's own actions with a delay results in the sensory information being processed similar to externally-generated, rather than self-generated, sensory input (e.g., Blakemore et al. 2000). Nevertheless, similar feedback delays have only limited effects on visuo-motor adaptation: despite changes in the rate of learning, the final extent of adaptation is only moderately affected with delays of 100-200 ms, and adaptation still occurs with delays as long as 5000 ms (Honda et al. 2012, Kitazawa et al. 1995, Tanaka et al. 2011). In other words, even in conditions where error assignment favors external factors, limb motor learning is relatively robust. For sensorimotor control of speech articulation, on the other hand, we have previously reported that auditory-motor adaptation to formant-shifted feedback is completely eliminated with delays of 100 ms or more (Max & Maffett 2015). Here, we test whether -- similar to visuomotor adaptation (e.g., Honda et al., 2012) -- the detrimental effects of delayed feedback signals can be minimized by first letting subjects habituate to the delay. Three groups of subjects completed an auditory-motor learning task in which a real-time vocal processor perturbed the subject's formant frequencies (i.e., speaker-specific resonance frequencies of the vocal tract) during the production of monosyllabic words (a manipulation known to induce an adaptive change in speech output). For the Control group, the formant-shifted feedback signal was delivered in real-time, but for the Habituation and Nonhabituation groups it was delayed by 75 ms. Prior to this adaptation task, each group completed 30 minutes of speaking tasks (word reading and picture naming): during these tasks, the Habituation group was already exposed to the 75 ms delay whereas the Nonhabituation group and Control group received only non-delayed feedback. All three groups showed adaptation in both the first and second formant. However, contrary to our hypotheses, the extent of adaptation was similar across all groups. Methodological differences between the present study and our previously published work may be key in understanding the effects of delayed feedback on sensorimotor adaptation in speech.

Disclosures: T. Mitsuya: None. **D. Shiller:** None. **L. Max:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.08/MM3

Topic: E.04. Voluntary Movements

Support: NIH grant R01DC13979

Hearing Research Inc.

Title: Responses to brief mid-utterance formant perturbations in the auditory feedback of ongoing speech

Authors: *I. RAHARJO[1,2], H. KOTHARE[1], J. F. HOUDE[1], S. NAGARAJAN[1]
[1]UC San Francisco, San Francisco, CA; [2]UC Berkeley, Berkeley, CA

Abstract: We can hear and monitor different aspects of our speech to make sure it is produced with appropriate loudness, prosody, and clear vowels to convey the intended meaning. If our feedback does not match our expectations, i.e. different from the intended speech, we tend to compensate to 'fix' the speech we produce. In a lab setting, this compensation response has been investigated using two different perturbation scenarios. In one scenario, the perturbation is introduced unexpectedly and temporarily. This causes speakers to immediately respond (short term response). Often, these perturbations are initiated at utterance onset, which has been done for pitch and formant alterations. Perturbations can also be initiated at mid-utterance which has only been looked at for pitch and amplitude alterations, but not for formants. Furthermore, it has been shown that responses to onset and mid-utterance perturbations differ in the case of pitch alterations. However, whether similar differences would be seen in formant perturbations are unknown. In another scenario, if this 'unexpected' perturbation is repeated over and over again, the perturbation becomes expected and an adaptation will likely take place (long term response). Here we explored three questions. First, we examined compensation responses to transient mid-utterance formant perturbation. Second, we compare these responses with responses to formant perturbations initiated at utterance onset. Third, we examined whether this compensation response is crucial for driving for formant adaptation. To examine formant perturbation responses, we applied a real-time shift of the first formant frequency (F1) to participants' utterance. The shifts performed were -100 Hz, +100 Hz, -50 Hz, and +50 Hz all equally distributed along with no perturbation trials. The shift is applied either for the whole trial at utterance onset or for only 400 ms after a jittered delay, while participants phonated the word 'head' (vowel /ɛ/) for 1.5 seconds. For the formant adaptation paradigm, we applied a constant F1 shift for several trials (preceded and followed by no perturbation trials) while participants phonated the word 'head' normally. This is repeated for each shift values that were used in the transient formant perturbation study.
We found that participants do compensate for mid-utterance formant perturbation. Their F1 response starts about 200ms after onset of formant perturbation. The peak magnitude of the response in both 100Hz and 50Hz (both polarities) is about 5 Hz. These results will be compared with the responses found from perturbations at utterance onset and to the adaptation response to consistent formant perturbation.

Disclosures: I. Raharjo: None. **H. Kothare:** None. **J.F. Houde:** None. **S. Nagarajan:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.09/MM4

Topic: E.04. Voluntary Movements

Support: NIH/NIDCD R01DC014510

Title: Speech auditory-motor learning: Are adaptation and de-adaptation similarly affected by practice schedule?

Authors: *K. S. KIM[1], *K. S. KIM[1], T. MITSUYA[1], L. MAX[1,2]
[1]Speech and Hearing Sci., Univ. of Washington, Seattle, WA; [2]Haskins Labs., New Haven, CT

Abstract: Sensorimotor adaptation plays a critical role in learning, maintaining, and refining voluntary movements. Surprisingly, few research efforts have been directed at uncovering the processes that occur when—after a period of sensorimotor adaptation—the feedback alteration or movement perturbation is removed (de-adaptation). The lack of research attention for de-adaptation may be related to the fact that subjects' behavioral responses gradually return to their pre-perturbation form, and that this change in behavior is often interpreted as an expected "forgetting" of newly learned internal models. This interpretation is typically based on the claim that de-adaptation is a faster process than adaptation. For some tasks, however, the rate of de-adaptation is not faster than the rate of initial adaptation even though the former involves re-adjusting movement parameters so that they once again account for sensorimotor transformations that previously had been experienced for many years. In addition, for limb movements, de-adaptation can take longer in children than in adults—if de-adaptation represents forgetting, this result would mean that children have better motor memory retention than adults. Instead, it may be more likely that de-adaptation itself represents new sensorimotor learning. Hence, de-adaptation may be better conceptualized as requiring an active re-learning of the appropriate sensorimotor maps rather than a process of forgetting. Addressing this question for auditory-motor learning in speech production, we investigated adaptation and de-adaptation as a function of the number of practice trials versus the amount of practice time during the corresponding phases of the experiment. For example, fewer trials performed per period of time in the post-exposure phase should lead to faster/increased de-adaptation if this phenomenon reflects mainly forgetting but to slower and decreased de-adaptation if the phenomenon reflects active sensorimotor re-learning of the non-perturbed environment. Forty adult subjects produced target words in pre-exposure (baseline), exposure (adaptation), and post-exposure (de-adaptation) phases of the experiment. During the exposure phase, all formants in the real-time auditory feedback signal were shifted 250 cents up. Participants were randomly assigned to four groups which differed in the rate at which trials were produced (either 18/min or 6/min) during the

adaptation and de-adaptation phases. Presentation of the findings will focus on the dissociation of effects due to the number of practice trials versus the amount of practice time and implications for speech motor learning and forgetting.

Disclosures: K.S. Kim: None. **T. Mitsuya:** None. **L. Max:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.10/MM5

Topic: E.04. Voluntary Movements

Title: High gamma neural responses dissociate between the acoustic and linguistic analysis of temporal speech structure

Authors: *G. B. COGAN[1], J. M. PEARSON[2], M. M. HAGLUND[1], S. R. SINHA[3], T. OVERATH[2]
[1]Dept. of Neurosurg., [2]Duke Inst. for Brain Sci., [3]Dept. of Neurol., Duke Univ., Durham, NC

Abstract: Speech perception entails the mapping of the acoustic waveform to its linguistic representation. For this mapping to succeed, the speech signal needs to be tracked across a large temporal range at high temporal precision in order to decode linguistic units (e.g. phonemes, syllables, words). Here we test how cortical processing of such temporal speech structure is modulated by higher-order linguistic analysis. To control the temporal scale of analysis, we used a novel sound-quilting algorithm that controls acoustic structure at different temporal scales; using fMRI, we recently showed that activity in human superior temporal sulcus (STS) increases as a function of temporal scale in an unfamiliar language (Overath et al., 2015). To control the linguistic content, we constructed speech quilts from both familiar and foreign languages. This ensures that any changes at the signal-acoustics level affect both languages identically, while manipulating the linguistic percept differently. Thus, neural responses that vary as a function of segment length but are shared or similar across the two languages suggest analysis at the signal-acoustics level, whereas neural responses that differ based on language familiarity imply the presence of linguistic processing. Here, we recorded electrocorticography (ECoG) from electrodes placed over left temporal or fronto-temporal lobes in three patients who were undergoing pre-surgical monitoring for pharmacologically resistant epilepsy. Patients listened to 6 s long English or Korean speech, quilted with 30 ms or 960 ms segment lengths. Electrodes with significant auditory responses were initially assessed via a permutation test between the 1 s time window following sound onset compared to the pre-stimulus baseline. Neural signals were filtered between 70 and 150 Hz (high gamma) and the results were Bonferroni corrected for multiple comparisons across electrodes. 20/124 electrodes demonstrated a significant auditory response. Within these electrodes, a follow-up analysis showed that sustained high gamma

responses throughout the 6 s sounds showed a main effect of segment length (30 vs. 960 ms) in 85% (17/20), a main effect of Language (English, Korean) in 60% (12/20), and an interaction in 50% of auditory electrodes (10/20). Specifically, electrodes that showed an interaction generally displayed a larger increase in high gamma power as a function of segment length in English than in Korean. These results suggest that high gamma neural responses are a potential neural mechanism for tracking speech-specific temporal structure. Ref: Overath T, McDermott JH, Zarate JM, Poeppel D (2015). *Nat Neurosci* **18**:903-911.

Disclosures: G.B. Cogan: None. **J.M. Pearson:** None. **M.M. Haglund:** None. **S.R. Sinha:** None. **T. Overath:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.11/MM6

Topic: E.04. Voluntary Movements

Support: NSF 1121147

Klarman Family Foundation

NSF GRFP

Title: Reciprocal connections between oral motor and agranular insular cortices mediate the control of consummatory behaviors

Authors: *L. M. AMARANTE[1], M. S. CAETANO[2], M. LAUBACH[1]
[1]American Univ., Washington, DC; [2]Univ. Federal Do ABC, Santo André, Brazil

Abstract: Traditional intracortical microstimulation (ICMS) studies in rodent have shown a role for the medial and lateral agranular frontal cortex (AGm - also known as M2 - and AGl, respectively) in oral facial motor responses. Additionally, studies have shown that these areas are connected with brainstem regions involved in the representation of jaw opening and closing and tongue protrusion. These studies did not assess cortico-cortical connections among these areas and did not report ICMS results in behaving animals. We performed ICMS in behaving and anesthetized rats and confirmed the motor output of the jaw/tongue region from stimulating in the AGm and AGl, as found in previous studies. A higher level of stimulation was necessary to evoke movements in AGm compared to AGl, suggesting that the AGl acts as a primary motor cortex and AGm acts as a secondary motor cortex for orofacial behaviors. ICMS in AGm of awake rats disrupted consummatory licking. Staining for the immediate early gene c-fos after microstimulation revealed anatomical connections between the AGm and the agranular insular / lateral orbitofrontal (AI/LO) cortex, an area traditionally implicated in gustatory responses.

These findings led us to investigate the anatomical and functional connections between these cortical regions. Retrograde tracing using Cholera Toxin Subunit B revealed direct anatomical connections from AI/LO to rostral AGm. Multi-electrode recordings in the two cortical areas during a consummatory licking task showed coherent rhythmic activity at the licking frequency. The recordings also found evidence for phase locking of LFPs to the lick cycle. These signals encoded relative reward value when rats licked for higher and lower concentrations of sucrose. Directed coherence analysis showed that lick-entrained activity across the two cortical areas was driven by activity in the rostral AGm. Multivariate analyses found evidence for correlation between the AI/LO and the rostral part of AGm, as well as anti-correlation between the rostral and caudal AGm. These findings, together with results from our ICMS and anatomical studies, suggest that reciprocal interactions between AI/LO and the rostral part of AGm may form a cortical circuit for the control of orofacial consummatory behaviors. We are currently investigating the interaction of these areas (AGm to AGl, and AGm to AI/LO) through using optogenetic perturbations with paired multi-electrode recordings in these areas.

Disclosures: L.M. Amarante: None. **M.S. Caetano:** None. **M. Laubach:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.12/MM7

Topic: E.04. Voluntary Movements

Support: NIH/NINDS R01 NS24328

NIH/OD/NINDS P40 ODO10996

NIH/NINDS P30 NS076405

NIH/NINDS T32 NS086749

Title: How primary is primary motor cortex for the control of vocalization?

Authors: *C. M. CERKEVICH[1,2,3,4], P. L. STRICK[1,2,3,4]
[1]Systems Neurosci. Inst., [2]Neurobio., [3]Univ. of Pittsburgh Brain Inst., [4]Ctr. for the Neural Basis of Cognition, Univ. Pittsburgh Sch. Med., Pittsburgh, PA

Abstract: Laryngeal muscles play a critical role in enabling vocalization in monkeys and humans. Yet, we know surprisingly little about the areas of the cerebral cortex that are involved in the descending control of these muscles. Here we used retrograde transneuronal transport of rabies virus to identify the cortical areas that are most directly connected to the motoneurons of laryngeal muscles in the macaque. This approach identified five cortical areas as the major origin

of output to laryngeal muscles. Two of these areas are on the lateral surface of the hemisphere and include the primary motor cortex (M1) and a region that overlaps portions of ventral area 6 (6V) and the motor proisocortex (ProM). Three of these areas are on the medial wall of the hemisphere and include the supplementary motor area (SMA), the rostral cingulate motor area (CMAr) and the ventral cingulate motor area (CMAv). We totaled the surface area of cerebral cortex that is the origin of descending control over laryngeal muscles. Then, we assessed the relative contribution of each motor area to laryngeal control. This analysis showed that M1 makes the single largest contribution to laryngeal control (~40%). The next largest output originates from two areas: 6V/ProM (~20%) and CMAr (~20%). In fact, taken together, the output from these two areas is equal to or greater than that from M1. Significantly smaller output originates from the SMA (~10%) and the CMAv (~6%). These results indicate that the descending control over laryngeal muscles originates from multiple cortical motor areas in the frontal lobe. M1 is the single largest source of cortical control over laryngeal muscles. Even so, the majority of the descending control originates from cortical areas outside of M1.

Disclosures: C.M. Cerkevich: None. **P.L. Strick:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.13/MM8

Topic: E.04. Voluntary Movements

Support: NIH NIDCD, DC014279

Title: Neural encoding of attended speech in primary and non-primary human cortices

Authors: *J. A. O'SULLIVAN[1,3], J. L. HERRERO[4,5], E. H. SMITH[2], G. M. MCKHANN[2], S. A. SHETH[2], A. D. MEHTA[6,5], N. MESGARANI[1,3]
[1]Electrical Engin., [2]Neurosurg., Columbia Univ., New York, NY; [3]Mortimer B. Zuckerman Mind Brain Behavior Inst., New York, NY; [4]The Feinstein Inst. For Med. Res., Great Neck, NY; [5]Hofstra Northwell Sch. of Med., Great Neck, NY; [6]The Feinstein Inst. for Med. Res., Great Neck, NY

Abstract: Humans posses the ability to segregate one speaker from another, even when there is no spatial separation between them. Invasive human neurophysiology studies have revealed a spectro-temporal representation of an attended speaker in non-primary auditory cortex (superior temporal gyrus; STG) that rapidly changes depending on the attentional focus of the listener. However, how such a representation emerges in STG is unknown. In addition, the representation of speech in primary auditory areas (Heschl's gyri; HG) remains unexplored due to their inaccessibility from the surface. Here, we used invasive depth electrodes and surface recordings

to investigate the neural encoding of attended speech along the auditory cortical pathway. Subjects listened to a male and female speaker, both in isolation (single-speaker; S-S), and mixed together (multi-speaker; M-S), with no spatial separation between them. To characterize the tuning properties of each electrode site, we obtained their spectro-temporal receptive field (STRF). Electrodes in HG tended to respond at short latencies (~50 to 150ms) and were narrowly tuned, with some electrodes responsive to speaker-specific features (e.g., the fundamental frequency of the male speaker). These electrode sites retained this speaker-selectivity, even when that speaker was not being attended to. Electrodes in STG responded at longer latencies (~150 to 300ms), were more broadly tuned, showed relatively little speaker-selectivity, and responded preferentially for the attended speaker. Comparing the STRFs from the S-S condition with the M-S condition, the tuning properties of HG electrodes remained similar, whereas electrodes in STG changed their tuning to enhance the attended speaker and suppress the unattended speaker. These results provide a descriptive account of how the acoustic features of attended and unattended speech are progressively processed along the auditory cortical pathway.

Disclosures: **J.A. O'Sullivan:** None. **J.L. Herrero:** None. **E.H. Smith:** None. **G.M. McKhann:** None. **S.A. Sheth:** None. **A.D. Mehta:** None. **N. Mesgarani:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.14/MM9

Topic: E.04. Voluntary Movements

Support: Japan Society for the Promotion of Science, No.26242040 for Scientific Research (A)

Japan Society for the Promotion of Science, No.16H06260 for Young Scientists (A)

Japan Society for the Promotion of Science, No.26861140 for Young Scientists (B)

Title: Decoding human speech articulation by multi-scale signals recorded from human sensory-motor cortex

Authors: *K. IBAYASHI[1], N. KUNII[1], T. MATSUO[2], Y. ISHISHIA[1], S. SHIMADA[1], K. KAWAI[3], N. SAITO[1]
[1]Dept. of Neurosurg., Univ. of Tokyo, Grad. Sch. of Med., Tokyo, Japan; [2]Tokyo Metropolitan Neurolog. Hosp., Tokyo, Japan; [3]Jichi Med. Univ., Shimotsuke, Japan

Abstract: Recent studies on Brain Computer Interface (BCI) has been proving its applicability in the clinical scene to restore human ability. Among the neural signals obtained from intracranial recordings, Single/Multi unit activity (SUA/MUA), Local field potential (LFP) and ECoG (Electrocorticography) are usually used as the input for the decoders used in BCI. Each of

these signals represent different scales of neuronal populations, and has been proven to be a good candidate as an input signal for BCIs especially for the control of prosthetic limbs. Our objective in this study was to evaluate the combined use of these multi-scale neural signals which were simultaneously recorded from the human ventral sensory-motor cortex (vSMC) while articulating Japanese phoneme. We recorded neural activities from subjects with refractory epilepsy, who underwent intracranial electrode placement for the purpose of evaluating the epileptic foci. 6 subjects whose vSMC was included in the potential peri-epileptogenic zone, had an electrode consisting of 6 microneedle combined with 3 macroelectrodes ("hybrid electrode") placed over the vSMC. The task was to pronounce a single Japanese phoneme displayed on a monitor. Articulation related neuronal activity was simultaneously recorded from the macroelectrodes and the microneedles. A standard expectation maximization clustering was performed to obtain SUAs by Offline Sorter® (Plexon Inc., Dallas, Texas, U.S.A.). LFPs and ECoG signals underwent time-frequency analysis to obtain spectral power for various frequency bands (Delta, Theta, Alpha, Beta, Low Gamma, High Gamma, Ultra-High Gamma). Spike firing frequency derived from SUAs and multi-frequency spectral power derived from LFP/ECoG were input as the feature vector. We constructed multiclass classifier using SLR (Sparse Logistic Regression) to decode spoken phonemes. Decoding accuracy for 5 spoken vowels were highest when feature from multiple signals were combined, which was 59% when averaged across subjects. The electrode location that performed best was compatible with tongue/lip area of human vSMC.
Although there are rooms for improvement, our results suggest that the hybrid electrode has the potential in providing consistent significant decoding accuracy by simultaneously recording multi scale neural activities. The less invasiveness in its implantation procedure and a possible longevity could be an advantage in future application for constructing speech-assisting BCI.

Disclosures: K. Ibayashi: None. **N. Kunii:** None. **T. Matsuo:** None. **Y. Ishishia:** None. **S. Shimada:** None. **K. Kawai:** None. **N. Saito:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.15/MM10

Topic: E.04. Voluntary Movements

Support: KAKENHI 26282165

Title: Oscillatory modulation during swallowing revealed by human electrocorticograms

Authors: *H. HASHIMOTO[1], M. HIRATA[2], S. KAMEDA[2], T. YANAGISAWA[2], S. OSHINO[1], H. KISHIMA[1], T. YOSHIMINE[2]

[1]Neurosurg., Osaka Univ., Suita-Shi, Japan; [2]Endowed Res. Dept. of Clin. Neuroengineering, Osaka University, Global Ctr. for Med. Engin. and Informatic, Suita-Shi, Japan

Abstract: Swallowing-related neural activity has been examined by non-invasive methods in human and there are several studies investigated by PET, TMS, NIRS, fMRI and MEG. We applied human electrocorticograms (ECoGs) to the analysis of swallowing-related neural activities. Four patients with intractable epilepsy participated in this study (4 females). In all cases, subdural electrodes were temporalily placed over the frontotemporal area. We injected 2 ml water bolus into their mouth by a syringe and asked them to swallow it at their own pace. Electroglottography (Laryngograph®) and a throat microphone were used to detect swallowing movement and timing. ECoG signals were measured using a 128-channel digital EEG system (EEG 2000; Nihon Koden, Japan) and digitized at a sampling rate of 1000 Hz. A 20 channel ECoG grid (5 × 4 contacts) which was placed over the caudolateral primary somatosensorymotor cortex was chosen for analysis. Fourier transformation and wavelet transformation were applied to ECoG signal. Power increase in the high frequency band (HFB, 76 - 100 Hz) appeared in the caudolateral pericentral gyrus, the frontal operculum and BA 43 during both swallowing preparation and execution. The power increase of BA 43 was strongest. Time-frequency plots of BA 43 demonstrated that high gamma band activity appeared from -1.0 s before swallowing corresponding to the preparation period and suddenly disappeared at 0.5 s after swallowing. At that time, there was no change in the low frequency band (LFB, 8-32 Hz), but LFB power increase was observed from 0.5 s after swallowing. High gamma band activities are more spatiotemporally focal than lower frequency activity and well reflect neural function. Therefore, high gamma band activity in BA 43 correlates with swallowing-related neural activities. High gamma band activity in BA 43 was observed within -1 to 0.5 s time interval which corresponded to swallowing preparation (-1 to 0 s) and oral phase (0 to 0.5 s). These phases are controlled voluntarily. High gamma band activity disappeared suddenly at 0.5 s and the activity transition from HFB to LFB was observed from 0.5s after swallowing. The involuntary swallowing starts from 0.5 s after swallowing execution and these periods are called pharyngeal and esophageal phase. This is the first reports that swallowing related high gamma activities were revealed. The switching of oscillatory modulation from HFB to LFB may reflect cerebral activity that converts voluntary swallowing into involuntary swallowing.

Disclosures: H. Hashimoto: None. **M. Hirata:** None. **S. Kameda:** None. **T. Yanagisawa:** None. **S. Oshino:** None. **H. Kishima:** None. **T. Yoshimine:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.16/MM11

Topic: E.04. Voluntary Movements

Support: NIH Grant DC 012502

CIHR MOP 137001

Title: Positive and negative BOLD contributions to speech production: An ICA approach

Authors: *V. L. GRACCO[1,2], H. OHASHI[1], N. BOURGUIGNON[3]
[1]Haskins Labs., New Haven, CT; [2]McGill Univ., Montreal, QC, Canada; [3]Ghent Univ., Ghent, Belgium

Abstract: Behavior reflects the engagement of distributed functional brain networks (FBN) that interact and overlap in reconfigurable ways. Here we focus on identifying the FBN engaged in the cognitive and sensorimotor processes underlying speech production. Our approach includes a detailed examination of the negative blood-oxygenation level dependent (BOLD) response (NBR) - a ubiquitous feature of all neural activity often associated with the default mode network. In contrast to the positive BOLD response (PBR) which has been associated with excitatory neural activity, the NBR appears to reflect suppressive operations. To understand the contribution of the PBR and NBR found in these networks during speech production, we used independent components analysis of fMRI data (ICA) to extract spatially distributed networks from global changes in the BOLD signal and examine their contribution to task manipulations and performance. We collected fMRI data from two speech production experiments in which speech was elicited under repetition conditions as well as word reading, picture naming and verb generation. We used these tasks to sample a range of control conditions from sensorimotor to cognitive. Speech production tasks engaged both task positive and task positive networks. General linear modeling and functional connectivity analyses were then applied to the ICA results to assess the functional contribution and interaction of the FBN supporting speech production. Several FBNs were identified in relation to specific tasks, together with several areas in which positive and negative BOLD signals overlap. Changes in functional connectivity within networks reflected flexible task positive and task negative interactions with task demands. Our results are consistent with a diverse and complex contribution of the PBR and NBR to speech production and to functional networks. These distributional characteristics of the positive and negative BOLD signals may reflect the consequence of excitatory and inhibitory interactions within and across these functional networks.

Disclosures: V.L. Gracco: None. **H. Ohashi:** None. **N. Bourguignon:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.17/MM12

Topic: E.04. Voluntary Movements

Support: 1DP20D008627-01

Title: Articulatory gesture encoding in human sensorimotor cortex during continuous speech production

Authors: *J. CHARTIER[1], G. K. ANUMANCHIPALLI[2], E. F. CHANG[2]
[1]Bioengineering, [2]Neurosurg., Univ. of California, San Francisco, San Francisco, CA

Abstract: To speak, we must coordinate over 100 muscles to precisely actuate our lips, jaw, tongue, larynx, and other vocal tract articulators. It is an extraordinary motor control feat, yet nearly all of us produce fluent speech. Our previous work focused on short consonant-vowel syllable production, and demonstrated that the human ventral sensorimotor cortex (vSMC) is functionally activated along somatotopic representations of articulators. However, natural continuous speech is far more complex and dynamic than single syllables because of co-articulation between adjacent segments and execution of motor plans over longer duration. To address this, we studied the encoding of kinematic properties in the human vSMC during natural sentence production. We recorded high-density intracranial electrocorticography signals, while speakers produced a set of sentences designed to cover all phonetic contexts in American English (MOCHA-TIMIT). We first developed a method to estimate vocal tract kinematic parameters from phonetic transcriptions and produced acoustics (acoustic-to-articulatory inversion). We then fit linear kinematic-trajectory models to each electrode using kinematic parameters to predict neural activity. We found single electrode encoding of dynamical representations of highly specific, coordinated out-and-back trajectories of articulators (e.g. tongue protrusion, lip closure, etc). Kinematic trajectories of electrodes clustered into four main categories differentiated by place of vocal tract constriction. Furthermore, electrodes in each trajectory category showed activations during the production of phonemes with similar places of articulation. Each trajectory category appeared to be spatially localized in the sensorimotor cortex. Lastly, the kinematic-trajectory model better explained electrode activity when compared against phoneme and single articulator representations. We have used natural continuous speech to demonstrate the neural representation of articulatory gestures in speech production.

Disclosures: J. Chartier: None. **G.K. Anumanchipalli:** None. **E.F. Chang:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.18/MM13

Topic: E.04. Voluntary Movements

Support: NIH Grant F32DC015708-02

Title: Cortical representation of articulatory gestures

Authors: *E. M. MUGLER[1], K. LIVESCU[4], M. C. TATE[2], J. W. TEMPLER[1], M. GOLDRICK[3], M. W. SLUTZKY[1]
[1]Neurol., [2]Neurolog. Surgery, [3]Linguistics, Northwestern Univ., Chicago, IL; [4]Toyota Technological Inst. at Chicago, Chicago, IL

Abstract: Speech is an important method of human communication. Despite decades of research, we do not completely understand the fundamental neural control of speech production. Current theories model speech production as a hierarchy, starting from sentences and phrases to words, syllables, speech sounds (phonemes) and - at the lowest level - the movements of speech articulator muscles to produce these sounds (articulatory gestures). We investigated the cortical representation of articulatory gestures and phonemes in speech motor, premotor, and inferior frontal cortices. We simultaneously recorded ECoG from M1v, PMv, and IFG (pars opercularis) and speech audio during single word utterances by human participants undergoing either functional mapping during awake craniotomies for resection of brain tumors or extra-operative monitoring for seizure localization. We manually labeled the onset of each phoneme and articulatory gesture. We analyzed the how cortical high gamma activity (70-300 Hz) varies with the context of phonemic and gestural events (i.e., co-articulation) in 9 subjects producing consonant-vowel-consonant words. The high gamma activity patterns across M1v and PMv did not change with position of the gesture within a word. In contrast, when aligned to phoneme onset, high gamma activity in M1v and PMv did vary with position within the word. We also used cortical activity to decode gestures and phonemes in each region. Combined M1v/PMv activity classified among all gestures with 65.6±5.5% accuracy (mean over subjects) and among all phonemes with 33.2±9.84% accuracy. Gestures were decoded significantly better than phonemes even after subtracting chance accuracy levels for each (paired t-test, $p = 5.2 \times 10^{-4}$). Our results indicate that primary motor and premotor areas more accurately represent gestures than phonemes. These findings suggest that the cortical control of speech production shares a common representation with that of other types of movement, such as arm and hand movements.

Disclosures: E.M. Mugler: None. **K. Livescu:** None. **M.C. Tate:** None. **J.W. Templer:** None. **M. Goldrick:** None. **M.W. Slutzky:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.19/MM14

Topic: E.04. Voluntary Movements

Support: NIDCD Grant DC014279

Title: Adaptive representation of noise and speech in human auditory cortex

Authors: *B. KHALIGHINEJAD[1,2], J. L. HERRERO[3,4], A. D. MEHTA[3,4], N. MESGARANI[2,1]
[1]Dept. Electrical Engineering, Columbia Univ., New York, NY; [2]Mortimer B. Zuckerman Mind Brain Behavior Institute, Columbia Univ., New York, NY; [3]Hofstra Northwell Sch. of Med., New York City, NY; [4]The Feinstein Inst. for Med. Res., New York City, NY

Abstract: Humans are experts at perceiving speech, even in the presence of interfering sound sources. Recent studies suggest that this perceptual ability may arise from a noise-invariant representation of speech in the auditory brain, resulting in several computational models to explain the robust encoding of stimulus. However, little is known about the dynamic properties of neural networks as the auditory system adapts to new noisy conditions.To understand the adaptation of auditory system during varying additive distortions, we recorded intracranial signals from four human subject who were implanted with high-density electrodes as a part of their clinical procedure. Subjects listened to continuous speech corrupted by additive background noise which changed randomly every three seconds to one of the four conditions of clean, bar, jet, and city noise.Analysis of the neural responses to the speech signal revealed an adaptation phase which lasted between 100 to 800 ms after the onset of noise change. This was also true for transitions from noisy to clean conditions. We investigated the encoding and representation of noise at each time point with respect to the transition time, and found that the representation of noise gradually degraded as subjects adapt to the new condition. Moreover, we examined the differences in spatial and correlation patterns between the adopted and non-adapted conditions, and found that correlation between activities of different brain regions increased during the adaptation phase.In addition, we show that current models of stimulus-response mapping fail to replicate the dynamic adaptation property of neural responses. To explore possible computations that could predict this dynamic response property, we implemented a cascade of Linear-Nonlinear models using a feed forward neural network. Incorporated a top-down adaptive mechanism that used the statistical structure of neural activation patterns allowed the model to replicate the temporal dynamic properties observed in the neural response at noise transitions. The proposed model thus offers insight into the biologically plausible computational mechanisms that can explain the robust speech perception observed in real-world hearing conditions.

Disclosures: B. Khalighinejad: None. **J.L. Herrero:** None. **A.D. Mehta:** None. **N. Mesgarani:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.20/MM15

Topic: E.04. Voluntary Movements

Support: H2020 Eurostars RapidMaps

Title: Real-time functional mapping assists electrical cortical stimulation to identify the eloquent cortex

Authors: *J. R. SWIFT[1,2], R. PRUECKL[3], C. KAPELLER[3], C. GUGER[3]
[1]Natl. Ctr. for Adaptive Neurotechnologies, Wadsworth Ctr., Albany, NY; [2]g.tec neurotechnologies USA inc, Rensselaer, NY; [3]Guger Technologies OG, Schiedlberg, Austria

Abstract: In several clinical scenarios, there is a need for associating cortical regions with their respective body functions. This is particularly relevant in the course of surgical treatment of patients who suffer from brain tumor or drug-resistant epilepsy. To minimize post-surgical functional deficiencies, it is essential to establish a functional map of involved cortical areas prior to the surgical intervention. Cortical areas covering important body functions such as movement, sensation, receptive and expressive language are commonly subsumed as “eloquent cortex.”
Electrical cortical stimulation (ECS) is considered the gold standard for identifying the eloquent cortex. During an ECS mapping session, electrical pulses are injected into the patient’s brain, leading to reversible excitation or inhibition of body functions. Relating the symptoms to the stimulation site allows establishing a functional brain map. This is however a tedious process for the patient, including the risk of pain or seizures.
An alternative to ECS is passive real-time functional mapping (RTFM), which is based on the analysis of brain signals invasively recorded via electrocorticography (ECoG). During an RTFM session, the patient performs certain tasks (movement, listening, speaking) while cortical activation in the High-Gamma Band (above 50 Hz) is extracted simultaneously for all ECoG electrodes. A functional map is then available within minutes without any risk for the patient.
In this study, we test whether a prevenient RTFM can assist ECS mapping to reduce stimulation time and risk. Four epilepsy patients underwent clinical ECS mapping and volunteered for an RTFM session. During the latter, all patients performed hand movement, tongue movement, and listened to words. We evaluated whether the RTFM-guided ECS outperforms random guidance. Additionally, the potential reduction of required stimulations was calculated.
On average, 52 electrode pairs were stimulated, where on average 18.8 electrodes related to motor, sensory, or language-related functions. The number of stimulations potentially obsolete due to RTFM guidance was 20.7 electrodes on average (reduction by 39.8%). Even with the reduced number of stimulations, all electrodes that had originally been revealed, were still identified. The RTFM significantly improved the ECS protocol for all four subjects ($p < 0.05$).
Compared to plain ECS, the proposed combined approach considerably reduces the overall mapping time and effort as RTFM indicates potential stimulation targets and therefore provides effective guidance. We are confident that the presented system improves functional mapping in clinical practice.

Disclosures: **J.R. Swift:** A. Employment/Salary (full or part-time):; g.tec neurotechnology USA inc. **R. Prueckl:** A. Employment/Salary (full or part-time):; guger technologies OG. **C.**

Kapeller: A. Employment/Salary (full or part-time):; guger technologies OG. **C. Guger:** A. Employment/Salary (full or part-time):; guger technologies OG.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.21/MM16

Topic: E.04. Voluntary Movements

Support: NSF Grant 1608140

BMBF 01GQ1602

Title: Reconstructing Mel-scaled spectrograms of speech from electrocorticography

Authors: *C. HERFF[1], G. D. JOHNSON[3], J. J. SHIH[5], D. J. KRUSIENSKI[4], T. SCHULTZ[2]
[1]Computer Sci., [2]Univ. of Bremen, Bremen, Germany; [4]Electrical & Computer Engin., [3]Old Dominion Univ., Norfolk, VA; [5]UC San Diego, San Diego, CA

Abstract: Remarkable progress in the decoding of speech aspects from neural recordings has been made in the last decade. The ultimate goal of restoring speech to severely disabled persons, however, is still far off. We use Electrocorticography (ECoG) to reconstruct the spectrogram of speech instead of classifying the speech process into discreet classes. In this study, ECoG activity was recorded during prompted speech from patients requiring surgical mapping for epilepsy treatment. While participant's spoke, ECoG and acoustic data were recorded simultaneously. Data was dissected into overlapping 50 ms long windows. The audio data was transferred into the frequency domain using Fast Fourier Transform. As human perception of frequencies is non-uniform, we scaled the spectrogram using the Mel-scale. To make the distribution of spectral power more Gaussian, we computed the logarithm of spectral coefficients. For the ECoG data, we extracted gamma power (70-170 Hz) for each channel and window. Brain process associated with speech production span a long duration prior to and after sound production. To capture this dynamic, neighboring feature vectors were stacked. Using regularized regression models, we show how the ECoG gamma activity can be directly mapped to the log mel-spectrograms. Our results show that the even simple regression approaches yield better than chance reconstruction of the speech spectrogram. This is an important step towards direct speech synthesis from brain activity.

Disclosures: C. Herff: None. **G.D. Johnson:** None. **J.J. Shih:** None. **D.J. Krusienski:** None. **T. Schultz:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.22/MM17

Topic: E.04. Voluntary Movements

Support: 1608140

Title: The neural correlates of lip movements during continuous speech for the development of speech neuroprosthetics

Authors: S. LESAJA[1], C. HERFF[3], G. JOHNSON[1], J. SHIH[4], T. SCHULTZ[3], *D. J. KRUSIENSKI[2]
[1]Biomed. Engin., [2]Electrical & Computer Engin., Old Dominion Univ., Norfolk, VA; [3]Computer Sci., Univ. of Bremen, Bremen, Germany; [4]UC San Diego, San Diego, CA

Abstract: Recent work has shown that it is possible to decode aspects of continuously-spoken speech from electrocorticographic (ECoG) signals recorded on the cortical surface. The ultimate objective is to develop a speech neuroprosthetic that can provide seamless, real-time synthesis of continuous speech directly from brain activity. The aim of this work is to investigate the neural correlates of speech-related lip movements from video recordings and to utilize this information in the development of improved decoding models. ECoG data have been collected from subjects undergoing clinical monitoring for epilepsy. The subjects performed a battery of speech tasks including modal and imagined continuous speech, as well as spontaneous modal and imagined speech via standard picture description and directed conversation tasks. The high gamma-band power (70-170 Hz) of ECoG was extracted and analyzed in conjunction with the recorded speech signals. From simultaneous video recordings, lip movement features (e.g., mouth opening height) were extracted using computer vision algorithms and correlated to the gamma-band activity at multiple time-lags. Preliminary analysis of the resulting spatio-temporal relationships between brain activity and lip movements will be presented.

Disclosures: **S. Lesaja:** None. **C. Herff:** None. **G. Johnson:** None. **J. Shih:** None. **T. Schultz:** None. **D.J. Krusienski:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.23/MM18

Topic: E.04. Voluntary Movements

Support: 'iConnect' project (grant ADV 320708)

'Language in interaction' project (NWO Gravitation grant 024.001.006)

Title: Decoding individual phonemes from ECoG neural responses using convolutional neural networks

Authors: *J. BEREZUTSKAYA, Z. V. FREUDENBURG, N. F. RAMSEY
Brain Ctr. Rudolf Magnus, Univ. of Utrecht, Utrecht, Netherlands

Abstract: Despite the large body of work on language decoding from the brain activity, it remains challenging to identify individual phonemes based on the neural responses. In the present study we used convolutional neural networks to decode classes of phonemes from the electrocorticography (ECoG) recordings. Four patients participated in an ECoG experiment. Each patient pronounced phonemes presented on the screen one by one. Four phonemes (/p/, /k/, /a/ and /u/) and a baseline rest condition were used. Microphone recordings of the patients pronouncing the phonemes were obtained simultaneously with the ECoG recordings. Each patient was implanted with a high density grid covering a mouth motor cortex. Gabor wavelet decomposition was used to extract high frequency band power signal from the raw ECoG data. Per patient the ECoG data were partitioned into trials. Each trial was defined as a [-0.5 - 1] s window around the voice onset (VO) of the phoneme. For the baseline condition the window was defined based on the cue onset. For decoding we constructed a 1D convolutional neural network (CNN) with one convolutional layer (32 filters) and a linear layer on top. The convolution was performed over time. Per patient the CNN was trained to predict the probability distribution over 5 classes (4 phonemes + rest) for each trial. In addition, for a number of patients we tested the CNN's capacity to identify these phonemes during a set of spoken words by applying the CNN using a sliding window over time. The CNN model showed significant decoding accuracy for the phoneme trials in all four patients: $a_1 = .8$, $a_2 = .54$, $a_3 = .81$ and $a_4 = .74$ (chance level = .2). The CNN decoding accuracy was comparable or better than the decoding accuracy obtained with a template match linear classifier ($\hat{a}_1 = .61$, $\hat{a}_2 = .36$, $\hat{a}_3 = .83$, $\hat{a}_4 = .61$). Per patient the decoding accuracy varied depending on the size and number of filters in the convolutional layer of the CNN. When tested on a continuous word pronunciation data, the CNN model reliably identified word periods vs rest. The exact phoneme decoding accuracy was moderate, due to presence of other phoneme classes the CNN had not been exposed to during the training. The CNN confused new phonemes with acoustically similar known classes: /b/ with /p/, /u:/ with /u/ and so on. In

the present study we trained a CNN to decode individual phonemes from the ECoG neural responses. In all patients the CNN model performed quite well on the neural data partitioned into phoneme trials. In addition, preliminary but promising results were obtained with identifying phonemes during a stream of spoken words. The results suggest that CNN analysis may facilitate decoding of spoken words from sensorimotor cortex.

Disclosures: J. Berezutskaya: None. **Z.V. Freudenburg:** None. **N.F. Ramsey:** None.

Poster

408. From Brain to Mouth: Oral Motor Speech Control

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 408.24/MM19

Topic: E.05. Brain-Machine Interface

Title: Topography of human motor speech area using externally recorded 12-20Hz beta peaks during articulatory movements and phoneme expression

Authors: *P. R. KENNEDY[1], T. LIMTOM[2], C. GAMBRELL[2], A. KIRILLOV[3]
[1]Neural Prosthetics, Neural Signals Inc, Duluth, GA; [2]Human Recording lab, Neural Signals Inc., Duluth, GA; [3]Neuroexplorer Inc, Houston, TX

Abstract: During our ongoing efforts to produce a near conversational rate speech prosthetic, we noticed that our locked-in subject produced 12-20 Hz beta peaks prior to the onset of covert speech (Sarmah and Kennedy 2013). Further studies in speaking adults demonstrated beta peaks at onset, offset and during inflection points in words or phrases (SFN Abstr. 2016). To investigate this further, we continue to record from the scalp over the motor speech area. This area was delineated by Bouchard and Chang using ECOG (2014), confirmed by functional MRI in PK, and further confirmed by cortical recording of single units related to articulatory movements, sensory responses and phonemes in PK. To determine if this topological map could be detected externally, we simultaneously record using four electrodes placed anterior and superior to the left ear. Recordings are made using CWE amplifiers at 200x gain and filtered between 1 Hz and 10 KHz. Data is archived on a Cygnus data recorder (at 10 KZ) and immediately digitized on a Neuralynx Cheetah software package using six continuous channel, four for the electrodes, one for speech and one for an event marker. Data is being analyzed using NEX (5.1 Neuroexplorer Inc. Texas, USA) with the assistance of Alex Kirillov, who wrote a script to assist in speeding up the analysis. To date, we have analyzed the six articulatory movements (jaw up and down, tongue protruded and retracted, cheeks pout out and retracted (grin). As these data show, these articulatory movements are differentially represented. With respect to the phonemes, the five vowel sounds and nine consonants are differentially displayed over this area, whether from the point of view of numbers of beta peaks or average amplitude of the beta peaks. We chose the vowels and consonants for analysis based on their articulatory

features such as fricatives, stops, nasals and so on. The aim of this project is to develop a simple speech prosthetic with a vocabulary restricted to 10 or 20 useful words distinguished on the basis of the timing of beta peak onsets, offsets and inflections points. In future studies, auditory feedback will be provided to the user so as to improve the consistency of the speech output and hence the beta peak patterns. These data suggest that a distributed pattern of beta peak production as well as the pattern found with a single electrode (SFN 2016) could be used to detect speech. This technique will be applied to locked-in subjects. The output from the recordings will be fed into a mobile phone loaded with an app that applies the beta detection and decoding software that drives words and phrases from the phone.

Disclosures: **P.R. Kennedy:** A. Employment/Salary (full or part-time):; Part time. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Neural Signals Inc.. **T. Limtom:** None. **C. Gambrell:** None. **A. Kirillov:** A. Employment/Salary (full or part-time):; Full Time, Neuroexplorer Inc.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.01/MM20

Topic: E.06. Posture and Gait

Support: JSPS KAKENHI Grant Number JP 15K16498

International academic conference travel subsidy from Sagami Women's University

Title: Improvement of cycling power output following transcranial direct current stimulation depends on the exercise duration

Authors: *S. SASADA[1], T. ENDOH[2], T. ISHII[3,4], K. KAWASHIMA[3], S. SATO[3], A. HAYASHI[3], T. KOMIYAMA[5,3]
[1]Dept. of Food and Nutr. Science, Sagami W, Sagamihara/Kanagawa, Japan; [2]Fac. of Develop. and Education, Uekusa Gakuen Univ., Chiba, Japan; [3]Fac. of Education, Grad. Sch. of Education, Chiba Univ., Chiba, Japan; [4]United of Grad. Sch. Education, Tokyo Gakugei Univ., Tokyo, Japan; [5]Dept. of Hlth. and Sports Sciences, Fac. of Education, Chiba Univ., Chiba City, Japan

Abstract: Fatigue is among the key factors determining practical motor performance in exercises of various durations, such as short and long distance categories in running, cycling, and skating. Recent studies showed that transcranial direct current stimulation (tDCS) to the leg area of the motor cortex could reduce fatigue and prolong time to exhaustion in submaximal cycling for ~6-

8 min, but it is unknown whether tDCS can improve motor performance for a wide range of exercise durations. Central fatigue, which is attributable to suboptimal central drive, is widely known to restrict motor output and gradually increase during both maximal and submaximal isometric force output. Therefore if central fatigue is involved with degradation of cycling performance, the effects of tDCS could differ depending on exercise duration. We therefore investigated how tDCS affects the power output in different durations of exhaustive cycling.We recruited 12 well-trained athletes. tDCS was applied with a stimulation electrode on the vertex and a reference electrode on the right forehead. tDCS was applied while the participants rested in a reclining chair. Three different stimulus polarities in stimulus electrode (i.e., anodal, cathodal, and sham) were tested. The stimulus intensity was 2 mA with 15-s ramp-up and -down periods. After 15 min of tDCS, the participants performed maximal effort sprint cycling for 8 s with a constant load (0.02 kp / kg body weight). The participants then performed an endurance cycling task involving submaximal, constant speed cycling until exhaustion. The load in this task started at 0.06 kp/ kg body weight and, after 5 min, increased in 0.5 kp steps every 3 min. In the endurance cycling task, anodal tDCS prolonged the time to exhaustion significantly more than cathodal or sham tDCS did. Anodal tDCS also resulted in significantly greater accumulative total power output than sham or cathodal tDCS did. In the sprint cycling task, neither anodal nor cathodal tDCS improved the peak or accumulative power relative to sham tDCS. Our findings showed that the effect of tDCS on cycling performance strongly depends on cycling duration and cycling type, with central fatigue progressing in different ways.

Disclosures: S. Sasada: None. **T. Endoh:** None. **T. Ishii:** None. **K. Kawashima:** None. **S. Sato:** None. **A. Hayashi:** None. **T. Komiyama:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.02/MM21

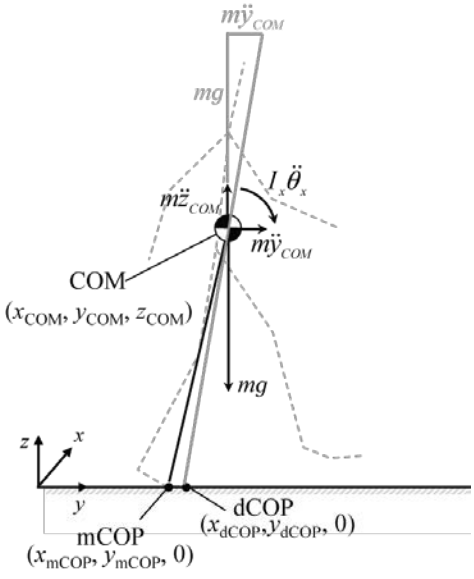
Topic: E.06. Posture and Gait

Title: Novel measure of dynamic balance during walking

Authors: J. YOO[1], T. YAMAGUCHI[2], M. SHINYA[3], M. MILOSEVIC[4], *K. MASANI[5,1]
[1]Inst. of Biomaterials and Biomed. Engin., Univ. of Toronto, Toronto, ON, Canada; [2]Grad. Sch. of Engin., Tohoku Univ., Sendai, Japan; [3]Hiroshima Univ., Higashi-Hiroshima, Hiroshima, Japan; [4]Grad. Sch. of Arts and Sci., Univ. of Tokyo, Tokyo, Japan; [5]Toronto Rehab Inst., Toronto, ON, Canada

Abstract: In the control system of humanoid robots, the key for maintaining postural stability during walking is to control the center of pressure under the entire body (COP). One successful strategy is to control the feet placement by aiming to place the measured COP (mCOP) on top of

the desired COP (dCOP). dCOP is defined as a virtual point on the ground, where the moment around the body center of mass (COM) becomes zero when dCOP and the measured COP (mCOP) coincide (i.e., a modification of the Zero-Moment Point concept used in the Honda humanoid robot)(Figure). Here we investigated whether this concept can be used to assess dynamic balance of human gait. We measured the mCOP and COM behavior (i.e., displacement, velocity and acceleration) of fourteen healthy male subjects during walking on an instrumented treadmill, under four conditions with different dynamic stabilities: (1) natural walking, (2) walking with rhythmic auditory stimulation, (3) walking while performing a cognitive task, and (4) walking with arms restrained. Then the dCOP was calculated using COM behavior according to Eq. 1. In all conditions, the trajectories of dCOP and mCOP were highly correlated during each step. Also, the distance between dCOP and mCOP (dCOP-mCOP) was relatively small on average. These results suggest that dCOP approximately follows mCOP. The small distance between dCOP and mCOP (dCOP-mCOP), which is proportional to the rotational acceleration of COM, was statistically different among the four conditions. The stride interval variability, which is an established measure of dynamic balance, also differed among the four conditions. Further, we found that the dCOP-mCOP was also highly correlated with the stride variability. These results supported that the dCOP-mCOP may cause gait instability, which results in larger stride variability. In conclusion, we suggest that a control strategy successfully used in humanoid robotics using dCOP may represent the dynamic balance of human gait, and may be used to assess people with deteriorated gait such as neurological patients and the elderly.



For y direction

Eq. 1 $$y_{dCOP} = y_{COM} - \frac{\ddot{y}_{COM}}{\ddot{z}_{COM} + g} z_{COM}$$

Eq. 2 $$y_{dCOP} - y_{mCOP} = -\frac{I_x \ddot{\theta}_x}{m(\ddot{z}_{COM} + g)}$$

Figure

Disclosures: **J. Yoo:** None. **T. Yamaguchi:** None. **M. Shinya:** None. **M. Milosevic:** None. **K. Masani:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.03/MM22

Topic: E.06. Posture and Gait

Support: JSPS KAKENHI Grant Number 15H05362

Title: Implicit manipulation of gait parameters by a visuomotor adaptation paradigm

Authors: ***K. YAMAMOTO**[1], T. IKEGAMI[2], M. HIRASHIMA[2]
[1]Grad. Sch. of Physical Educ., Natl. Inst. of Fitness and Sports In Kanoya, Kagoshima, Japan; [2]Natl. Inst. of Information and Communications Technol., Suita City, Osaka, Japan

Abstract: A central issue in human gait analysis is to manipulate gait parameters like gait speed, step width, or foot angle, and assess its effects on joint and muscle loads during gait. Most previous studies have investigated this issue by asking participants to explicitly modify these gait parameters. However, such explicit intervention often leads to some exaggerated or unnatural behavior at unspecified parts of the body. More critically, how much to modify the gait parameters is not properly controlled but depends on participants. Thus, explicit modification may not be appropriate to systematically manipulate gait parameters in human gait analysis. Here we propose a new methodology to use a visuomotor adaptation paradigm, which allows participants to implicitly modify gait parameters during gait. We constructed an elaborate experimental setup in which participants walk on a treadmill while viewing real-time visual feedback of their own movements through an avatar skeleton, which is scaled to each participant's body size and displayed on the front screen. The visual feedback was presented as a computer-generated motion of the avatar so that visuomotor discrepancy between actual and virtual body can be artificially added. In this study, we specifically examined whether participants can implicitly modify the "step width" (i.e., mediolateral distance between the heels in double support phase). Two parallel rails were displayed as a virtual walking road on the screen. The distance between the rails was fixed as a natural step width for each participant, which was recorded in advance without visual feedback. Five healthy males (22.2±1.48 yr) were asked to walk while viewing the avatar's motion so that each step of the avatar contacted on the corresponding rail (i.e., left step on left rail). During outward (inward) adaptation trials, in the first 5 minutes, we gradually added a visuomotor discrepancy whereby the avatar's step width was displayed to be smaller (larger) than the actual one by the participant. Then the visuomotor discrepancy was gradually returned to the original level, zero in the next 5 minutes. Our participants significantly increased or decreased their actual step width according to the

magnitude of discrepancy in outward ($F(4,2) = 19.7$, $p < 0.001$) or inward ($F(4,2) = 34.0$, $p < 0.001$) adaptation trials, respectively. Importantly, all participants were unaware of the visuomotor discrepancy applied on the step width. The result suggests that gradual visuomotor adaptation technique can implicitly guide human behavior, and thus is useful to systematically manipulate participants' gait kinematics without any deliberate motions.

Disclosures: K. Yamamoto: None. **T. Ikegami:** None. **M. Hirashima:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); 3D Incorpolated.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.04/NN1

Topic: E.06. Posture and Gait

Support: HHS/NIDILRR grant H133E070013

Title: Biomechanical effects of differential limb-loading demands in the fore-aft direction on relative interlimb propulsion symmetry during walking

Authors: *A. NAIDU[1], C. P. HURT[2], D. A. BROWN[3]
[1]Rehabil. Sci., Univ. of Alabama At Birmingham, Birmingham, AL; [2]Dept. of Physical therapy, [3]Physical Therapy, Univ. of Alabama at Birmingham, Birmingham, AL

Abstract: **PURPOSE:** Hemiparesis following stroke impairs paretic (P) limb-loading dynamics and propulsive-force generation during walking. By integrating a split-belt treadmill with a robot-assistive device, we have developed a novel method to differentially load one limb relative, specifically in the for-aft direction during walking. Our long-term goal is to develop a treadmill-based training paradigm to improve P limb propulsive-force generation and participation during walking. Hence, the purpose of this study is to measure the relative interlimb propulsion symmetry in nonimpaired individuals, engaged in walking in our novel robotic, split-belt, environment under differential limb-loading demands in the fore-aft direction. Results from these studies will be compared with a similar paradigm in individuals poststroke.
HYPOTHESIS: In nonimpaired individuals, a single limb working against greater fore-aft, limb-loading demands will comparatively increase its propulsive-force during walking due to greater proprioceptive load-feedback integration, causing interlimb-propulsion (IP) asymmetry.
METHODS: Nonimpaired individuals walked at tied-belt (TB) speeds (1m/s) within the robotic interface at different trunk inclination angles (IA) correlated with low, mid and high force effort, against the interface's pelvic mechanism. To induce differential limb-loading participants walked at the same IA's with belts split in a 2:1 split-belt (SB) speed ratio. Steady state kinetic

(propulsion) and kinematic data (stride length) for all trials was compared.
RESULTS/CONCLUSION: Data of 7 participants (Male:2, Mean age 28 ± 5 years) was analyzed at 2 TB and 4 SB differential conditions at low and high force effort, respectively. No difference in interlimb-propulsion impulse (i.e. time integral of propulsion) during TB conditions was seen. However, an 11% symmetrical increase was noted from low to high effort TB trials, respectively. During the SB-1 conditions (left=1 Right=0.5m/s), IP asymmetry increased from 2% at low effort to 17% at high force effort respectively, with greater propulsion by the right (slower moving) leg. IP asymmetry was also noted during the SB-2 conditions (left=1 Right=1.5m/s), with greater propulsion by the left (slower moving) leg from 0.3% at low effort to 11% at high force effort, respectively.
IMPLICATIONS: Results from this work will enhance mechanistic understanding of how differential fore-aft loading demands can influence individual-limb neuromuscular force responses during walking, in nonimpaired individuals. Potentially, these results can also be used to test new split-belt training paradigms for people poststroke.

Disclosures: A. Naidu: None. **C.P. Hurt:** None. **D.A. Brown:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); consultant to HDT robotics, co-inventor.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.05/NN2

Topic: E.06. Posture and Gait

Title: Balance and muscle activation patterns during sit-to-stand across four initial foot positions

Authors: *W. JEON[1], D. GUPTA[1], J. FENNELL[2], J. L. JENSEN[1], L. GRIFFIN[1]
[1]Kinesiology and Hlth. Educ., The Univ. of Texas At Austin, Austin, TX; [2]Univ. of Melbourne, Melbourne, Australia

Abstract: *Introduction:* A posterior foot position before the onset of sit-to-stand (STS) has been reported to have several kinetic advantages. Previous studies, however, have not assessed the ideal initial foot position (IFP) and its relationship to balance during STS. The purpose of this study was to investigate leg muscle activation patterns in 4 different symmetric IFP's (neutral; both knees at 90° flexion, and one-third, two-thirds and three-thirds of each participant's foot length posterior to neutral) and to examine the effects of IFP on maintaining balance after standing. *Methods:* EMG activity of 4 leg muscles (rectus femoris (RF), gastrocnemius (GAST), biceps femoris (BF), and gluteus maximus (GM)) during STS was measured in 10 healthy individuals and normalized to the EMG recorded during maximum voluntary contraction of each muscle. The standard deviation (SD) of the ground reaction force (GRF) (antero-posterior and

lateral) for 1 second after completing fully upright posture in 4 different IFP's was used to measure balance during the stabilization phase. *Results:* On average, EMG activity of RF and GAST increased as IFP moved posteriorly from neutral to three-thirds of the individual's foot length (21% to 35% and 11 to 21% EMGmax respectively), while BF and GM decreased (19 to 11% and 21 to 10% EMGmax respectively). The antero-posterior SD of GRF was the lowest at the posterior two-thirds IFP and was highest at the neutral IFP ($p < 0.05$). There was no difference in the lateral SD of GRF across the 4 IFP's. *Conclusion:* While there is a trade-off of leg extensor and thigh extensor muscles with changes in IFP, the posterior two-thirds foot length IFP is the most stable for maintaining balance during the stabilization phase before quiet standing.

Disclosures: W. Jeon: None. **D. Gupta:** None. **J. Fennell:** None. **J.L. Jensen:** None. **L. Griffin:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.06/NN3

Topic: E.06. Posture and Gait

Title: Movement dynamics associated with response strategies in running

Authors: *J. L. MCDONNELL[1], J. C. MIZELLE[2], T. R. DERRICK[4], S. MEARDON[3]
[2]Kinesiology, [3]Physical Therapy, [1]East Carolina Univ., Greenville, NC; [4]Kinesiology, Iowa State Univ., Ames, IA

Abstract: Introduction: Single subject analysis of movement strategies adopted throughout the acquisition of a motor tasks is imperative in understanding dynamic task performance. Specific footwear features, such as midsole hardness, elicit adaptive movement dynamics, allowing examination of response strategies. Degree of adaptation falls on a continuum between a mechanical response (MR) in which landing forces increase with harder midsoles and a neuromuscular response (NMR) in which forces are neutralized. Kinematics, kinetics, and individual characteristics associated with response strategies have not been evaluated in the context of internal model stability. The purpose of this study was to examine responses to footwear changes and potential explanatory factors.
Methods: 40 participants were categorized as either a mechanical (MR) or neuromuscular (NMR) responder based on the change in the rate of ankle joint contact force loading (ROL) across 3 midsole hardness conditions during running. MRs exhibited consistent reduction of the both the peak ankle joint contact force and ROL, with a softer midsole. Inversely, NMR did not display a change in force or ROL across footwear conditions. Group differences in variables thought to influence response strategy included: age, height, mass, sex, running pace, volume

and experience, injury history, foot mobility, joint and leg stiffness and somatosensation.
Results: 29 MR demonstrated an increased ROL with increased midsole hardness and 11 NMR exhibited no significant change in ROL across midsole hardness conditions. Of the demographic variables examined, only years of running experience differed between groups (MR: 8±3.5, NMR: 4.8 ±2.8). Further, both MR and NMR exhibited increased ankle stiffness as midsole hardness increased while only the NMR displayed decreased knee stiffness with increasing midsole hardness.
Discussion: Adopted motor strategies for different shoe material may be related to internal model stability. Repetitive execution of behavior generates persistent changes in kinematic and neurologic measures. The adherence to a movement pattern regardless of varying conditions suggests a stable internal model in which feedback and feedforward mechanisms maintain intended strategy of execution. Response strategies observed here may reflect the dynamical process of motor skill learning, where sensory-motor coupling can update an existing internal model to increase stability.
Conclusions: MR possessing significantly more years of experience, may reflect the influence of time on adaptive responses. Further analysis of jerk, a well-established indicator of motor adaptation is warranted.

Disclosures: J.L. McDonnell: None. **J.C. Mizelle:** None. **T.R. Derrick:** None. **S. Meardon:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); New Balance.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.07/NN4

Topic: E.06. Posture and Gait

Title: Contribution of lower limb joint movement to symmetrization of step length in split-belt locomotion

Authors: *K. HIRATA[1], T. KOKUBUN[2], H. YOKOYAMA[3], T. MIYAZAWA[4], K. KUBOTA[1], M. SONOO[1], H. HANAWA[1], N. KANEMURA[2]
[1]Grad. Course of Hlth. and Social Services, Grad. Sch. of Saitama Prefectural Univ., Koshigaya-Shi, Japan; [2]Dept. of Hlth. and Social Services, Saitama Prefectural Univ., Koshigaya-Shi, Japan; [3]The Univ. of Tokyo, Tokyo, Japan; [4]Ageo Futatsumiya Clin., Ageoshi, Japan

Abstract: [Introduction] Most studies on the split-belt walking paradigm, in which two belts are driven independently of each other, were utilized as models to demonstrate the adaptability of human bipedal locomotion. Predictive feedforward mechanism was proved by the interlimb parameters, such as step length and double support time, changed slowly during adaptation

process, eventually acquiring symmetry. However, kinetic contribution to the symmetrization of step length has not yet been clarified. Step length is defined by the cosine theorem using the length of leading leg and its limb angle, and the length of trailing leg and its limb angle at heel strike. This study aims to investigate the contribution to step length about limb angle and lower limb joint angles (hip and knee joints). [Methods] Ten young adults walked on split-belt treadmill in symmetric and asymmetric conditions. In these conditions, we investigated the changes in limb angle, hip angle, knee angle, and step length at heel strike using Vicon motion capture system. All parameters were corrected three periods, which in symmetric conditions (baseline period), asymmetric conditions before adaptation (early adaptation period), and after adaptation (late adaptation period). The ratio from three periods were analyzed using ANOVA with repeated measures. Post-hoc analyses were performed using Tukey's honest significant difference test when the ANOVA yielded significant results. The statistical significance level for all measures was set as $p < .05$. [Results] Hip angle at heel strike on one side was not significantly different between the baseline and the late adaptation periods similar to the step length. In contrast, hip angle at heel strike on the other side, limb angles and knee joints showed significant differences between the baseline and the others periods ($p < .05$). [Discussion] In the present study, the symmetrization of the step length occurred as results of the limb angle of the fast leg increased and that of the slow leg decreased at heel strike on the fast side. Similarly, these of the limb angles on the slow leg decreased and those on the fast leg increased at heel strike on the slow side. However the limb angle was asymmetry at heel strike, the hip flexion angle was symmetry. Namely, the hip flexion movement could be predictively controlled similar to step length and double support time. Then, it is possible that the lower limb length could be extended to minimize the vertical movement of the center of gravity at the time of grounding by reducing the knee flexion movement derived from the hamstrings. In conclusion, the dynamic strategy of predictive feedforward control during locomotion was suggested.

Disclosures: K. Hirata: None. **T. Kokubun:** None. **H. Yokoyama:** None. **T. Miyazawa:** None. **K. Kubota:** None. **M. Sonoo:** None. **H. Hanawa:** None. **N. Kanemura:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.08/NN5

Topic: E.06. Posture and Gait

Support: NIH Grant T32HD007490

Title: Retention of locomotor learning: Effects of practice duration and washout periods

Authors: *J. E. GALGIANI, S. M. MORTON
Physical Therapy, Univ. of Delaware, Newark, DE

Abstract: Locomotor adaptation has been studied extensively using the split-belt treadmill paradigm (i.e. where the two feet are driven at different speeds). The cerebellum has been shown to be an essential brain region responsible for the initial learning of the new split-belt walking pattern, but it is less clear what areas are responsible for its long-term retention. Likewise, although it has been postulated that shorter practice durations and use of a 'washout' period should lead to relatively reduced retention, evidence for this is not sufficient. Here, we exposed two groups of young, healthy adults to split-belt treadmill walking with the belt speeds at a 3:1 ratio. The SHORT group adapted for 5 minutes on Day 1 followed immediately by a 5 minute washout period in which they walked with the belts tied. The LONG group adapted for 15 minutes on Day 1 and had no washout period. We examined retention of the learned split-belt walking pattern (changes in step length symmetry) 24 hours later, when both groups returned to the lab and performed a 15 minute bout of split-belt walking. We hypothesized that on Day 2, the SHORT group would demonstrate reduced recall, relearning and overall retention of the walking pattern learned the previous day. Though the performance of the LONG group was better than the SHORT group at the end of adaptation on Day 1, surprisingly, we found that neither the shorter period of walking nor the completion of a washout immediately after learning reduced how well subjects in the SHORT group remembered the next day. That is, neither immediate recall, nor relearning, nor overall retention was different between the SHORT and LONG groups. This suggests that split-belt learning in healthy individuals is quite robust, and that it is possible to see retention with very little practice. Additionally, these data may warrant re-examination of the previous idea that retention is dependent on practice duration.

Disclosures: J.E. Galgiani: None. **S.M. Morton:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.09/NN6

Topic: E.06. Posture and Gait

Support: NHMRC Fellowship APP1002190

NHMRC Program Grant APP1091302

Title: Is it "pain, no gain" rather than "no pain, no gain" for motor learning?

Authors: *P. W. HODGES[1], S. E. SALOMONI[2], L. J. BOUYER[3]
[1]Univ. Queensland, Brisbane, Australia; [2]The Univ. of Queensland, Brisbane, Australia; [3]Rehabil. Dept- CIRRIS, Univ. Laval Fac Med., Quebec, QC, Canada

Abstract: Here we questioned the commonly accepted assumption that ***pain*** should be endured to make ***gains*** *in motor performance* - that is, “no pain, no gain”. The contrasting view is that pain may affect the ability to learn new motor skills and this is critical to resolve as it has implications for training sport skills in the presence of pain or developing movement strategies to overcome pain in rehabilitation. Limited evidence in humans shows contrasting results from major to no interference of pain on learning. Some also report compromised learning on subsequent training of the same task after pain resolution. We aimed to resolve this debate by studying the impact of task-related pain on learning during treadmill walking perturbed by a force field that pulls the leg up and forward. This requires compensation by early activation of knee flexor muscles to successfully overcome the perturbation and prevent falling. The task was performed on 2 consecutive days. On Day 1, electrical stimuli over the posterior thigh elicited pain with an intensity that was induced in a manner that was proportional to the degree of knee flexor muscle activity (Pain group N = 13; Control N = 14). The task was practiced again on Day 2 in the absence of pain. Kinematics of the lower limb were recorded with a three-dimensional motion analysis system (Vicon, USA) using a standard marker set. The novel force perturbation applied to the leg on Day 1 led to increased foot velocity compared to baseline walking and required several strides before an adaptation was learned to overcome the perturbation. Compared to pain-free walking, task-related pain reduced the overall performance (greater foot velocity) and led to a different adaptation of gait, with less optimal features such as a longer swing phase duration. This solution results in suboptimal performance, with greater active torque and potential for higher metabolic costs. This less ideal gait pattern was repeated when the task was practiced again 24 hours after pain exposure, despite the absence of pain. Our results show that pain does not prevent learning, but can modify the solution used to learn a new task. This modified solution is likely suboptimal and is retained during re-exposure to the same task, despite absence of pain. Our results indicate that the presence of pain limits gains in motor performance, contradicting the old adage of “no pain, no gain”. As far as learning a new motor skill and recovery of function are concerned, training through pain may not be ideal after all.

Disclosures: P.W. Hodges: None. **S.E. Salomoni:** None. **L.J. Bouyer:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.10/NN7

Topic: E.06. Posture and Gait

Support: JSPS KAKENHI Grant Number JP17K01479

Title: Proximal-to-distal segmental sequence of toppling observed during human quiet standing

Authors: *S. SASAGAWA[1], A. IMURA[2]
[1]Dept. of Human Sciences, Kanagawa Univ., Yokohama, Kanagawa, Japan; [2]Dept. of Hlth. Promotion Sci., Tokyo Metropolitan Univ., Tokyo, Japan

Abstract: Recent studies have shown that all major leg joints (i.e., ankle, knee, and hip) have significant contributions to the control of human standing balance (e.g., Yamamoto et al. 2015). However, what kinematic patterns exist among these joints is still unclear. In the present study, by focusing on "micro falls," which are characterized by destabilizing rise in velocity of the body's center of mass (CoM) and occur frequently during quiet standing (Loram et al. 2005), and by using time-locked averaging technique, we examined the kinematic patterns among the three leg joints. Ten healthy young participants were required to stand quietly on a force platform for 60-s with eyes open. The anterior-posterior (AP) position of the center of foot-pressure (CoP) was measured by a force platform. At the same time, the ankle, knee, and hip joint angles in the sagittal plane were calculated from the three-dimensional Cartesian coordinates of reflective markers attached to the characteristic points of the participants' body. By low-pass filtering the CoP data, the time-series of the AP position of the CoM was estimated (Caron et al. 1997). The CoM velocity and the CoM acceleration were obtained by single and double numerical differentiation of the position data, respectively. In the following analysis, we identified CoM velocity maxima (negative-going zero-crossing of CoM acceleration) for the averaging points at the center of the micro falls and averaged the kinematic data 1.0 s either side of these points to highlight the signal features. As a result, we found a proximal-to-distal sequence of forward toppling of the body segments during the micro falls; i.e., at first the hip starts flexing, then the knee extending, and finally the ankle dorsiflexing. With respect to the angular velocity maximum, the knee and ankle lags the hip by approximately 400 ms and 700 ms, respectively. To understand neural mechanisms behind such sequential toppling, further investigations focusing on muscle activities and/or joint torques are needed.

Disclosures: S. Sasagawa: None. **A. Imura:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.11/NN8

Topic: E.06. Posture and Gait

Title: Effects of motor fatigue on walking stability during concurrent cognitive challenges

Authors: *P.-C. KAO, M. A. PIERRO, K. BOORAS
Physical Therapy, Univ. of Massachusetts Lowell, Lowell, MA

Abstract: Cognitive-motor interference, a negative influence on the performance of one or both tasks, is manifested when simultaneously performing a cognitive task and a motor task. It has been shown that motor fatigue reduces neuromuscular performance such as a temporary decline in the ability of generating required force level. However, little is known about the effects of motor fatigue on the cognitive performance and more importantly, on the cognitive-motor dual-tasking performance, an important capability during our daily lives. The purpose of this study is to investigate how motor fatigue affects people's cognitive performance (e.g., information processing, working memory), motor performance (i.e., walking), and the performance in cognitive-motor dual tasking. We recorded kinematic data as subjects walked on a treadmill at 2.8 mph under three different conditions: walking only, while receiving the Paced Auditory Serial Addition Test (PASAT), and a modified color-word Stroop test before and after a submaximal muscle fatigue exercise session that includes leg presses, calf and toe raises. We quantified walking stability by computing dynamic margins of stability (MOS), gait variability, and short-term local divergence exponent (LDE) of the trunk motion. For dynamic MOS, we calculated the distances between the "velocity-adjusted" center of mass position and boundaries of base of support at heel strikes. The preliminary data of seven healthy young subjects show that subjects demonstrated significantly greater local instability ($p<0.05$) in the mediolateral trunk motion after the muscle fatigue session compared to the baseline. Subjects had significantly greater mean MOS_{AP} and mean MOS_{ML} after the muscle fatigue session by walking with greater step length and width compared to the baseline. In addition, subjects had significantly greater variability in the trunk motion, MOS, lower-extremity joint angels and spatiotemporal gait measures after muscle fatigue. During the dual-task conditions, subjects walked with similar local stability level but significantly less variability of knee and hip joint angles as well as less MOS variability compared to walking only, suggesting that subjects may try to control their foot placement and joint kinematics during walking. For the cognitive performance, there are no significant differences in the number of errors during the PASAT or the modified Stroop test before and after muscle fatigue. These preliminary data suggest that motor fatigue does not affect cognitive performance but motor performance under the cognitive-motor dual-task conditions in healthy young individuals.

Disclosures: P. Kao: None. **M.A. Pierro:** None. **K. Booras:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.12/NN9

Topic: E.06. Posture and Gait

Support: NSF 1535036

GAANN program P200A150050

Title: Locomotor learning "in the wild": Motorized shoes can induce split-belt-like adaptation over ground

Authors: *Y. AUCIE[1], X. ZHANG[2], R. SARGENT[2], G. TORRES OVIEDO[1]
[1]Bioengineering, Univ. of Pittsburgh, Pittsburgh, PA; [2]The Robotics Institute, Carnegie Mellon Univ., Pittsburgh, PA

Abstract: There is a clinical interest to correct step length asymmetry post-stroke (i.e., limp) because it impairs patients' mobility. Promising studies have shown that stroke survivors recover gait symmetry after walking on a split-belt treadmill that augments their step asymmetry by moving the legs at different speeds. However, the transfer of gait improvements to over ground walking is limited. We hypothesize that gait improvements would be more general if we could induce locomotor learning by augmenting step asymmetry like split-belt treadmills while walking over ground. Thus, we developed motorized shoes, called Nimbus, that can move the legs at different speeds while walking over ground. In this study, we determined if the Nimbus shoes could induce similar gait adaptation effects to those observed on the split-belt treadmill. Thus, we compared walking kinematics between subjects wearing the Nimbus shoes on a regular treadmill (n=8) vs. subjects wearing regular shoes on a split-belt treadmill (n=9). Both groups experienced an adaptation period when the legs move at different speeds (3:1 speed ratio). Positions from the ankle and the hip were collected bilaterally and used to compute step length, step position, and step time asymmetry, which are known to adapt during split-belt walking. These parameters were used to contrast between groups 1) the extent of adaptation (i.e., changes in gait from early to late adaptation) and 2) the magnitude of after-effects (i.e., changes in gait before and after the adaptation period). We found that subjects in both groups adapted their gait as shown by the changes in behavior from early to late adaptation. However, the extent of adaptation was smaller in the Nimbus than split-belt group in step position (step asymmetry $p=0.104$; step position: 0.036; step time $p=0.863$) likely due to the smaller velocity differences in the Nimbus than split- belt group (Step velocity: $p<0.001$). Importantly, subjects from both groups had after-effects in all parameters except for step time in the split-belt group, as shown by the significant differences in behavior before and after the adaptation phase (step asymmetry: Nimbus $p<0.001$, split-belt $p<0.001$; step position: Nimbus $p<0.001$, split-belt $p<0.001$; and step time: Nimbus $p=0.003$; split-belt $p=0.172$). This indicates longer lasting after-effects in gait timing in the nimbus group than the split-belt group. In sum, our results demonstrate that the Nimbus shoes are portable devices that can induce error-based locomotor learning comparable to split-belt walking but outside the laboratory, which holds the great promise of improving patients' gait during real- life situations beyond the clinic.

Disclosures: **Y. Aucie:** None. **X. Zhang:** None. **R. Sargent:** None. **G. Torres Oviedo:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.13/NN10

Topic: E.06. Posture and Gait

Support: NIH 5K01NS092785-02

Title: Can split-belt treadmill walking be explained with a reflex-based model?

Authors: *S. SONG[1], Y. AUCIE[2], G. TORRES-OVIEDO[3]
[1]Robotics Inst., Carnegie Mellon Univ., Pittsburgh, PA; [2]Bioengineering, [3]Dept. of Bioengineering, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Gait adaptation on split-belt treadmills provides insights on the underlying control structure for walking. For example, observations on infants and adults walking on split-belt treadmills with various speed configurations have led to a consensus that the locomotion controller consists of separate functional networks for each leg and for different locomotion modes (e.g., forward vs. backward walking). However, most of the interpretations of these experiments are based on an assumption that the spinal motor circuits are governed by central pattern generators (CPGs). Here, we investigate the possibility that humans adapt their gait without CPGs. In other words, we evaluated the extent to which human gait adaptation on split-belt treadmills moving the legs at different speeds can be reproduced in simulation by a spinal-reflex-based neuromechanical model, which consists of a network of spinal reflexes mediated by supraspinal control without CPGs. Our results show that the reflex-based neuromechanical model can successfully generate stable split-belt walking with one leg moving at 1.5 m/s and the other one at 0.5 m/s. Moreover, our preliminary results show that, when the reflex control parameters are optimized for minimum metabolic consumption, the model reproduces most of the stepping features observed in human split-belt treadmill walking. Specifically, we performed a one-sample t-test to find significant differences between the gait features of nine healthy subjects and those produced by our model and found that both the subjects and the model converged to the same step-position ($p=0.25$), step-time ($p=0.010$) and step-velocity ($p=0.056$). Interestingly, we found differences in the step length asymmetry reached by the simulation and the experimental results ($p<0.001$), suggesting that metabolic consumption may not be the only factor optimized in humans. We are currently investigating the effect of optimizing for different costs, including metabolic energy, muscle fatigue, and gait asymmetry, to explore the physiological basis of human gait adaptations upon sustained changes in the walking environment imposed by the split-belt treadmill. Once we identify the cost function driving locomotor learning, we will further investigate the contributions of individual reflex pathways in

the gait adaptation of the model. The findings will allow us to augment gait rehabilitation with devices such as the split-belt treadmill.

Disclosures: S. Song: None. **Y. Aucie:** None. **G. Torres-Oviedo:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.14/NN11

Topic: E.06. Posture and Gait

Title: Interference in locomotor adaptation

Authors: *A. SALATIELLO[1], G. TORRES-OVIEDO[2]
[1]Bioengineering, [2]Dept. of Bioengineering, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Split-belt treadmill walking, in which legs move at different speeds, can be used to improve patients' mobility by correcting their gait asymmetry (e.g. Reisman et al 2013). For this strategy to be effective it is necessary to maximize the *retention* of motor memories acquired during the training. In order to do so, it is important to know how motor memories learned within the same environment influence each other. In this study we specifically tested whether learning two locomotor patterns counteracting equal and opposite perturbations is possible or instead the memories *interfere* with one another. To this end we studied unimpaired subjects' ability to counteract the same perturbation twice after either experiencing an opposite perturbation in-between (interference group, n=8) or walking without any perturbation (savings group, n=8). Critically, unlike prior work (Malone et al 2011), we removed the opposing perturbation gradually to reduce the experienced errors known to reinforce the motor memory initially learned (Herzfeld et al. 2014). As a measure of error, we used step length asymmetry. We compared across groups 1) the change in initial error that subjects experienced when the perturbation was introduced, 2) the change in steady state value reached and 3) the percent change in adaptation rate. We found that while both groups had similar initial change in errors (and thus where similarly perturbed, p=0.69), and similar change in steady state values (indicating a comparable ability in facing the perturbation at steady state, p=0.11), the dynamics of adaptation were significantly different. In fact, the interference group re-adapted 38.27% slower (p=0.035, 95% bootstrap CI [3.52%, 80.99%]), whereas the savings group re-adapted as fast as during the first exposure. In sum, our results indicate that the memory of adapted walking patterns is subject to *interference* and that this memory can be reinforced by the errors experienced during de-adaptation. These findings can inform the design of more effective rehabilitation techniques to counteract step length asymmetry in stroke survivors.

Disclosures: A. Salatiello: None. **G. Torres-Oviedo:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.15/NN12

Topic: E.06. Posture and Gait

Support: FAPESP Grant 2015/13100-9

FAPESP Grant 2016/23571-1

FAPESP Grant 2013/02322-5

FAPESP Grant 2013/01050-1

Title: Treadmill versus overground body weight support training in stroke patients: Effects on coordination variability during walking

Authors: *M. L. CELESTINO[1,2], A. M. F. BARELA[1], G. L. GAMA[1], J. A. BARELA[1,3], R. E. A. VAN EMMERIK[2]
[1]Inst. de Ciências da Atividade Física e Esporte, Univ. Cruzeiro Do Sul, Sao Paulo, Brazil; [2]Dept. of Kinesiology, Univ. of Massachusetts, Amherst, MA; [3]Dept. de Educação Física, Univ. Estadual Paulista, Rio Claro, Brazil

Abstract: Gait training with body weight support (BWS) system is commonly performed while walking on a treadmill during gait rehabilitation of patients with stroke. It is currently unknown how BWS training on differently surfaces (treadmill versus overground) affects coordination during walking. The aim of this study was to assess the effects of two different training protocols with BWS on gait intra-limb coordination variability of patients with stroke. Twenty-eight patients were recruited and were randomly assigned to two training groups: overground group (OG) and treadmill group (TM). They performed two evaluations of free overground walking: T0 (pre-training) and T1 (post-training). During the evaluations, a video system of movement capture (VICON) was used to obtain the kinematic data and reconstruct hip and knee joint angles for both paretic and non-paretic limbs. After the T0 evaluation, both groups participated in six weeks of training with BWS, three sessions per week (18 sessions). Vector coding was used to quantify intra-limb coordination between hip and knee joints of paretic and non-paretic sides. The key dependent variable was the coordination variability between stride cycles, derived from the mean phase angle between hip and knee joint angular changes. For both the paretic and non-paretic sides, there existed significant interactions between training group and evaluation, with the OG group increasing coordination variability after training (T0 to T1) and the TM group decreasing variability after training (p's $< .0001$). A similar interaction was found for the non-paretic limb during the swing phase, with higher variability on T1 for the OG and lower

variability on T1 for the TM group. No differences were observed for the swing phase on the paretic side. We conclude that the training with BWS on different surfaces impacts gait coordination differently; training overground with BWS improves gait adaptability in both paretic and non-paretic limbs, especially during the stance phase, as shown by increased coordinative variability. In contrast, training on a treadmill with BWS reduces coordinative variability and possibly gait adaptability during free overground walking.

Disclosures: M.L. Celestino: None. **A.M.F. Barela:** None. **G.L. Gama:** None. **J.A. Barela:** None. **R.E.A. van Emmerik:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.16/NN13

Topic: E.06. Posture and Gait

Support: NIH NICHD Grant R21HD088342

Title: Learning and generalization of locomotor skills acquired during a virtual obstacle negotiation task

Authors: *A. KIM, J. M. FINLEY
USC, Los Angeles, CA

Abstract: Obstacle negotiation is an essential skill for everyday locomotor performance. Recent studies have demonstrated that obstacle negotiation can be trained in a goal-oriented manner by providing people with auditory feedback of errors between actual and desired levels of foot clearance. Although the performance of this skill can be improved within a single day of training, it remains to be seen how this type of locomotor skill is retained and generalized to over-ground walking. We developed a novel virtual obstacle negotiation task to assess how locomotor skills are learned in fully-immersive virtual environments and how this learning generalizes to obstacle negotiation in the real world. This study involved two visits on consecutive days. Day 1 began with an over-ground baseline where participants negotiated a single physical obstacle ten times (PRE). They then completed a treadmill-based, skill acquisition phase which involved stepping over 120 virtual obstacles viewed through a head-mounted display (Oculus Rift DK2). Participants were instructed to minimize the clearance between the foot and the obstacles during the crossing step. During acquisition, participants were provided with three types of auditory performance feedback: a pleasant sound when foot clearance was within a target range of 0-2 cm, an error sound whose frequency scaled with foot height when clearance was greater than 2 cm, and a failure sound following collisions with the obstacle. Generalization was assessed by repeating the over-ground baseline procedure (POST).

On Day 2, participants revisited the lab and completed one retention block of 40 virtual obstacles with no auditory feedback. We then assessed over-ground retention in the same manner as the generalization trials on the previous day. There was a significant improvement in performance during the acquisition period as participants reduced foot clearance from 6.0 ± 2.6 cm in Block 1 to 3.2±1.4 cm at the end of Block 3 on Day 1. We observed some forgetting on Day 2 as initial foot clearance increased 1.3±1.0 cm in the retention trial compared to the end of Day 1. However, compared to the initial foot clearance on Block 1, the initial foot clearance on Day 2 (4.5±1.7 cm) was lower. The improvements observed in the virtual environment also transferred to the real world. Clearance was reduced from 13.8 ± 2.9 cm during PRE to 9.6 ±1.2 cm during POST and some of this improvement was retained on Day 2 (10.7 ± 1.3 cm). Overall, these results extend previous work by demonstrating that obstacle negotiation skills can be learned and retained in VR, and also demonstrating that locomotor skills acquired in VR can be transferred to the real world.

Disclosures: A. Kim: None. **J.M. Finley:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.17/NN14

Topic: E.06. Posture and Gait

Support: NIH Grant K12HD073945

James Zumberge Individual Research Award from the University of Southern California

Title: Regulation of whole-body angular momentum during adaptive locomotor learning

Authors: *S. PARK[1], J. M. FINLEY[2]
[1]Biokinesiology and Physical Therapy, [2]USC, Los Angeles, CA

Abstract: One of the primary challenges of bipedal locomotion is to transport our bodies through the environment without falling. Although we typically achieve this goal through the use of symmetric walking patterns, it is unclear if we actively vary asymmetry to maintain stability when adjusting to novel demands during walking. Here, we explore this possibility in the context of adaptation to walking on a split-belt treadmill. As people adjust to walking with the belts moving at different speeds, they gradually change their foot placement and timing to take steps of equal length. Although the acquisition of symmetry has been described as an error-based learning process, it is also possible that adaptation stems a desire to control whole-body dynamics to maintain balance. Therefore, we determined how whole-body dynamics are

regulated during split-belt adaptation, and how this regulation is affected when stability is augmented by allowing participants to hold on to a handrail.
Participants adapted to walking on a split-belt treadmill at a 3:1 belt speed ratio and we measured whole body dynamics using a full-body marker set. To characterize changes in whole-body balance during adaptation, we computed the angular momentum about the center of mass (CoM) and quantified how peak-to-peak angular momentum and integrated angular momentum were modified over the course of adaptation. We hypothesized that both measures of angular momentum would be reduced over the course of adaptation in parallel with changes in step length asymmetry. We further hypothesized that providing an external source of stability by holding on to handrails would reduce the range of whole-body angular momentum during both early and late adaptation.
We found that the peak-to-peak range of the whole-body angular momentum for each stride was increased during early adaptation compared to baseline and then decreased as people reduced step length asymmetry over the course of adaptation. Counter to our initial hypothesis, changes in the peak-to-peak range of angular momentum were consistent across stability conditions. These results indicate that regulation of the whole-body angular momentum was indeed perturbed by the imposed asymmetry during early adaptation and improved during adaptation. However, since there were no significant differences in adaptation or momentum control across stability conditions, control of whole body rotational dynamics does not appear to be a sensitive measure of the change in fall risk across stability conditions.

Disclosures: S. Park: None. **J.M. Finley:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.18/NN15

Topic: E.06. Posture and Gait

Support: NSERC (Canada)

Title: The influence of objects in the peri-personal space on standing sway

Authors: M. A. BRYANTON[1], *K. K. FENRICH[2], J. E. MISIASZEK[2]
[1]Occup. Therapy, [2]Fac. of Rehabil. Med., Univ. of Alberta, Edmonton, AB, Canada

Abstract: Background: The proximity of an object within an individual's peri-personal space has been previously documented to impact postural sway in healthy young adults. While some researchers suggest that improved postural stability occurs due increased optic flow, which provides greater online visual feedback of body sway, others suggest that a restriction of sway and shifting of center of pressure (COP) away from the object occurs to prevent contact with the

object regardless of the availability of vision. Aim: The purpose of this investigation was to evaluate both the effect of object proximity and positioning on COP sway characteristics with and without visual feedback. Methods: Participants were asked to stand quietly on a foam pad placed on a force platform, with feet shoulder width apart, for 90s. A cardboard wall (i.e. spatial restriction device (SRD)) adjusted to neck height was placed either directly in front, or to the side (left and right) of the participant, at two distances (10 and 20 cm relative to the 1st distal phalange for front placement or lateral edge of the foot for left/right placement). These 6 locations of the SRD were tested with eyes open (EO) and eyes closed (EC) for a total of 12 experimental conditions. Two additional trials without the SRD were also performed to characterize baseline sway characteristics with EO or EC. Conditions were presented randomly across participants. Results: When vision was provided, the mean COP position tended to shift away from the SRD location. In addition, when the SRD was placed directly in front, preliminary results showed a marked reduction in COP sway area (EOF10: 56.8±22.5%; EOF20: 52.1±13.0%) and velocity (EOF10: 22.9±17.0%; EOF20: 19.4±15.2%). These pronounced effects on sway area and velocity were not apparent when the SRD was placed to either side of the participant. The placement of the SRD had no effect on sway parameters during EC trials, despite the participants being aware of the location of the SRD prior to closing their eyes. Discussion: These results indicate that objects in the peri-personal space reduce postural sway only when vision is provided. This suggests that either 1) the threat of contact with the object is not a critical influence in the control of standing sway, or 2) visual feedback is required to accurately estimate the location of objects in space. Regardless, these findings indicate that sway is influenced by the presence of objects in the peri-personal space, which may represent a simple means of introducing a progressive, adaptive training paradigm for standing balance control.

Disclosures: M.A. Bryanton: None. **K.K. Fenrich:** None. **J.E. Misiaszek:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.19/NN16

Topic: E.06. Posture and Gait

Support: University of New Mexico OVPR Grant

University of New Mexico OFAC Grant

Title: Improvement of dynamic balance control by an exercise program

Authors: *D. SHIBATA
Univ. of New Mexico, Albuquerque, NM

Abstract: Central processing of multi-sensory feedback and motor commands responsible for force production are critical for balance control. An exercise program designed to realign spinal curvature was recently developed, but its effect on balance control is unknown. This study examined changes in center of gravity (CoG) sway on stable and unstable surfaces after completing the exercise program. Subjects (n = 27) were randomly assigned into one of three groups: exercise on a cylinder-shaped tube (StretchPole, LPN Corporation, Nagoya, Japan, Ex-SP group, n = 9), exercise on a flat surface (Ex-FS group, n = 9), and a control group that rested on a flat surface (n = 9). The exercise program consisted of three preparatory positions and seven small motions, and the session lasted approximately 15 mins. Subjects in the Ex-SP group performed the exercise program while lying on the cylinder-shaped tube, while those in the Ex-FS group performed the same exercise program lying on a flat surface. Subjects in the control group lay supine on a flat surface for 15 mins. Each subject's pre- and post-test CoG sway was measured while standing on a balance system platform (Biodex Medical Systems, Shirley, NY, USA) under a static and dynamic condition. Subjects were instructed to stand still and the platform was held stationary (e.g., no tilt) during the static condition. During the dynamic condition the platform was allowed to tilt in response to changes of CoG and subjects were instructed to maintain the platform in a horizontal position to the best of their ability. Subjects performed 5 trials of each test condition, for 20 s each. Two 2 □3 mixed-model ANOVAs were used to examine changes in average CoG sway and group differences. The hypotheses of this study were: a) the decrease in Ex-SP group CoG sway would be significantly greater than in the other two groups, and b) the decrease in CoG sway during the static platform test condition would be significantly larger than during the dynamic platform test condition.
There were no significant differences during the stable platform test condition. A significant interaction was observed during the dynamic platform test condition, and *post-hoc* analyses revealed that CoG sway significantly decreased in the Ex-SP group ($p < 0.05$), but there were no differences in the other two groups. These findings indicate that CoG sway significantly improved when completing the exercise program on a cylinder-shaped tube, but only when the standing surface was unstable. It is speculated that performing the exercise program on a cylinder-shaped tube might enhance central processing of sensory feedback, as well as the motor commands responsible for force production in balance control.

Disclosures: D. Shibata: None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.20/NN17

Topic: E.06. Posture and Gait

Title: Quantifying the non-linear properties of center of pressure patterns during different running styles

Authors: *C. D. BOWERSOCK, S. MORRISON, D. RUSSELL
Old Dominion Univ., Norfolk, VA

Abstract: Running is a popular mode of exercise used to meet recommended physical activity levels and sustain a healthy lifestyle. In order to better understand the benefits and disadvantages of different running styles, the majority of studies have focused on lower limb kinematics and/or kinetics. A less common approach has been to assess changes in center of pressure (COP) progression over successive steps during running. Given the current trend to utilize different running styles to decrease injury risk, consideration of how the COP structure may change during different running styles should be made. The aim of this study was to quantify the dynamic structure of three different running styles and identify the unique signatures of each. Specifically, we examined differences in basic spatiotemporal measures and COP measures using linear and non-linear analysis. Nineteen young active individuals without prior lower extremity injuries participated in the study. Each individual ran at a standardized speed under three different running conditions, namely; 1) preferred running style, 2) forefoot strike pattern and, 3) rearfoot strike pattern. All persons performed the task on a Zebris instrumented treadmill with an inbuilt pressure plate. A single 5-minute trial was performed for each condition with the COP and spatiotemporal measures recorded continuously for the trial duration. Standard linear analyses were used to quantify COP displacements, step length, stride time and cadence. Non-linear analyses included detrended fluctuation analysis (DFA) and approximate entropy (ApEn) to quantify the COP structure. The results revealed several differences between the three running styles. The forefoot condition was characterized by shorter COP path length, decreased stance times and increased cadence compared to the other two running patterns. The ApEn analysis revealed inherent differences between the three running patterns with regards to their pattern of regularity while no differences were observed for the DFA analysis. The DFA results produced values between 0.5 and 1 across the 3 running conditions, suggesting persistence in the COP structure irrespective of the point of initial contact. Several differences in the COP dynamics were observed between running styles using ApEn measures. In particular, the ApEn results revealed differences in the structure of the COP patterns, implying each running style possess its own unique COP signature. Together, the findings of this study highlight that assessing COP patterns during running provide insight to the dynamics underlying different running styles.

Disclosures: C.D. Bowersock: None. **S. Morrison:** None. **D. Russell:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.21/NN18

Topic: E.06. Posture and Gait

Title: Saccadic eye movements influence postural sway after fatiguing and stretching

Authors: *J. M. HONDZINSKI, M. A. YEOMANS, A. G. NELSON, M. J. MACLELLAN
Kinesiology, Sch. of Kinesiology; Louisiana State Univ., Baton Rouge, LA

Abstract: The purpose of this study was to better understand postural control declines that stem from muscular fatigue and provide insight into a possible mechanism to improve these declines. Fatigue can decrease muscle force and increase sway during quiet stance to diminish postural control compared to a non-fatigued state. In contrast, by performing saccadic eye movements during quiet stance, people can attenuate postural sway thus possibly improve postural control. We reasoned that saccades may also attenuate sway accompanying fatigued states and chose to test this in the present study. Barefooted young adults were asked to stand still on a force plate and either stare at a fixation point (FIX) or to perform saccadic eye movements (SAC) by viewing targets appearing alternately at 1 Hz on left and right sides of a computer screen at a 20 degree visual angle. Subjects performed 6 trials (3 FIX and 3 SAC) under non-fatigued (NF), plantar- and dorsi-flexed stretched (S), and plantar- and dorsi-flexed fatigued (F) conditions with either a wide or narrow base of support (BOS). We included S as a secondary condition, as it can decrease muscle force without fatigue. Performance of NF trials preceded S trials which preceded F trials. S involved rounds of passive unweighted dorsiflexion and plantarflexion immediately before performance of every two trials. F involved toe rises on a step until range of motion decreased below 75% or subjects could do no more prior to each trial. Analyses performed on several sway measures for the center of pressure revealed the following. When using a narrow BOS during FIX, S frequently attenuated sway and F often intensified or equaled sway compared to NF conditions, while use of saccades often attenuated sway. Outcomes varied by measures so that changes occurring with different conditions influenced sway in different directions. Moreover, the condition influences were not as evident with a wide BOS. Results of the present study match previous work which showed fatigue can deteriorate standing sway, yet contrast others which indicated that stretching also deteriorates standing sway. Differences for the latter can be explained by unilateral verses bilateral stance and/or the muscles stretched, as opposing muscles were stretched in the current study. However, changes in somatosensory feedback remain a candidate for altered postural control after fatiguing and stretching. These data also support the use of saccades to increase the external focus of attention to improve postural control. We conclude that saccades may improve postural control thus standing balance in populations susceptible to muscle fatigue.

Disclosures: J.M. Hondzinski: None. **M.A. Yeomans:** None. **A.G. Nelson:** None. **M.J. MacLellan:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.22/NN19

Topic: E.06. Posture and Gait

Title: Analysis of kinematic synergy in arm swing during gai

Authors: *T. FUJINO, N. KANEMURA, T. KOKUBUN, K. KUBOTA, K. HIRATA, H. HANAWA, A. KOBAYASHI, K. TAKAYANAGI
Saitama Prefectural Univ., Koshigaya-Shi, Japan

Abstract: Kinematic synergy allows the movement of redundant joint systems in a coordinated manner. It is known that foot placement is stabilized by kinematic synergy of the lower limb joints during gait. Moreover, arm swing contributes to gait stability and energy efficiency. However, kinematic synergy of the upper limb joints is little understood. The purpose of this study is to investigate kinematic synergy in the arm swing during gait and to clarify on the basis of what synergy, is the arm swing during gait controlled. Eight healthy young male volunteers participated in this study. Arm swing movement was measured during self-selected gait velocity on a treadmill. Joint motions were measured using motion capture system and trunk segment angle on frontal and horizontal plane and elevation angles of body segments (upper arm, lower arm) on sagittal plane were calculated from measured motion. Each angular velocity was calculated by the first derivative of segment angles. In order to extract kinematic synergy, uncontrolled manifold (UCM) analysis was performed on important performance variables. Hand placements and velocities in anterior-posterior (AP) and superior-inferior (SI) directions were used as performance variables in the UCM analysis. The angle and angular velocities were set to elemental variables, and "good variance" (VUCM), which does not affect performance variables, and "bad variance" (VORT), which affects task performance variables, were calculated. ΔV, which is an index of synergy, was obtained from VUCM and VORT. The more positive ΔV, the stronger the synergy. Negative values suggest the absence of a stabilizing synergy. In the case where hand placement was taken as a performance variable, ΔV was negative value(AP:-0.39±0.05,SI:-0.47±0.15). There was no kinematic synergy that stabilizes step-by-step variations in both AP and SI directions. In contrast, ΔV was positive value(AP:0.08±0.02,SI:0.43±0.11), when using hand velocity as a performance variable, in both directions, the existence of kinematic synergy was observed. In arm swing during gait, it was suggested that there is a cooperative structure to stabilize variations in hand velocity. Hand velocity strongly reflect angular velocity of the upper extremity. Since the angular velocity of the upper limb is affected by the angular momentum of other segments, such as the trunk and lower limbs, the kinematic synergy restrains variance of the angular momentum, which may contribute to gait stability. These preliminary findings show that hand velocities during gait is controlled

more coordinative than hand placements and suggested possibility to more reflect control by central nervous system.

Disclosures: T. Fujino: None. **N. Kanemura:** None. **T. Kokubun:** None. **K. Kubota:** None. **K. Hirata:** None. **H. Hanawa:** None. **A. Kobayashi:** None. **K. Takayanagi:** None.

Poster

409. Posture and Gait: Kinematics, Muscle Activity, Exercise, and Fatigue

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 409.23/NN20

Topic: E.06. Posture and Gait

Title: Automatic unsupervised analysis of motor function by deep learning

Authors: *B. OMMER[1], B. BRATTOLI[1], U. BÜCHLER[1], A.-S. WAHL[2], M. E. SCHWAB[3]
[1]Heidelberg Univ., Heidelberg, Germany; [2]Brain Res. Institute, Univ. and ETH Zurich, Zuerich, Switzerland; [3]Univ. Zurich, Zurich, Switzerland

Abstract: Motor behavior analysis provides a crucial non-invasive and easily accessible diagnostic tool for biomedical research. A detailed analysis of posture changes during skilled motor tasks can reveal distinct functional deficits and their restoration during recovery. Our specific scenario is based on a neuroscientific study of rodents recovering from a large sensorimotor cortex stroke and skilled forelimb grasping is being recorded. Given large amounts of unlabeled videos that are recorded during such long-term studies, we seek an approach that captures fine-grained details of posture and its change during rehabilitation without costly manual supervision.
Therefore, we have developed a deep learning approach that utilizes self-supervision to train a convolutional neural network to analyze motor function. The underlying posture and behavior representations are automatically learned by having the convolutional neural network watch a large number of unlabeled grasping videos. Achieving this goal depends on the following fundamental contributions: (i) automatic limb detection based on a fully convolutional network that is initialized solely using motion information, (ii) a novel self-supervised training of long short term memory networks using only temporal permutation yields a detailed representation of behavior, and (iii) back-propagation of this sequence representation also improves the description of individual postures.
We have established a novel test dataset with expert annotations for evaluation of fine-grained behavior analysis. Moreover, we demonstrate the generality of our approach by successfully applying it to self-supervised learning of human posture on standard benchmark datasets.

Disclosures: B. Ommer: None. **B. Brattoli:** None. **U. Büchler:** None. **A. Wahl:** None. **M.E. Schwab:** None.

Poster

410. Posture and Gait: Animal Models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 410.01/NN21

Topic: E.06. Posture and Gait

Support: NIH NINDS NS054894,

NIH NINDS NS072651

Title: Contributions of pelvic roll to weight-supported locomotion in the neonatally spinalized rat

Authors: *J. VANLOOZEN, S. F. GISZTER
Neurobio. and Anat., Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: Rats spinalized at T9/10 as neonates (P5/6) show recovery of hindlimb alternation as adults (NTX).Around 20% of animals show autonomous weight-supporting locomotion without therapeutic intervention. Utilizing robot-assisted treadmill-based rehabilitation based at the pelvis, some NTX animals increase the amount of weight-supported stepping across a 30-day training period. Previous work has demonstrated that expansion of trunk motor cortex is critical for NTX animals who learn to weight-support using the pelvic rehabilitation paradigm (Oza, 2015). Lesioning of trunk motor cortex in weight-supporting NTX animals reduces their ability to take weight-supported steps (Giszter, 2008).
Trunk motor cortex is likely of particular importance in spinal animals undergoing locomotor rehabilitation because it serves as a mechanical link between cortically-controlled upper trunk muscles and spinally-controlled areas under the lesion site. Further investigating the effects of locomotor training on pelvic roll can reveal mechanisms and effects of trunk motor cortex expansion in weight-supporting spinal animals. Kinematics were recorded from animals throughout robot-assisted treadmill training at the hip and ankle using OPTOTRAK 3020. Pelvic roll, pitch, yaw and translation and foot position across the step-cycle were used to correlate changes in biomechanics to changes in weight-supported stepping. Roll control seems especially important.
In animals that transition from low levels of weight-supported stepping (<50% WSS) to higher levels of weight-support (>50% WSS) (TWS NTX) we reduced pelvic roll precedes expansion of the hind-limb task-space; together these changes enable development of weight-supported stepping in NTX animals. In NTX animals beginning training at a high-level of weight-support pelvic roll remains stable with training, with minor limb representation expansion. Animals beginning training at a low level of weight-support that fail to increase weight-supported stepping with training do not stabilize pelvic roll either early or late in training. In some the hindlimb task-space was expanded, albeit non-functionally, without a reduction in pelvic roll so

that animals were unable to take weight-supported steps.
These data suggest that control of pelvic roll via trunk musculature is a critical component of successful locomotor rehabilitation in the NTX rat. Reduction in pelvic roll occured prior to expansion of the hindlimb limb task space in the TWS NTX, and suggests that rehabilitation of trunk control balance motor action components may be an important first step in locomotor rehabilitation of spinal animals.

Disclosures: J. Vanloozen: None. **S.F. Giszter:** None.

Poster

410. Posture and Gait: Animal Models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 410.02/NN22

Topic: E.06. Posture and Gait

Support: NIH Grant NS086973

Title: Adaptations of neural control to mediolateral perturbations of the patella

Authors: *F. BARROSO, C. ALESSANDRO, T. SANDERCOCK, M. TRESCH
Northwestern Univ., Chicago, IL

Abstract: Research on motor control is usually done considering task level variables, for example, by assessing how the central nervous system (CNS) produce joint torques that successfully accomplish desired behavioral goals such as stable locomotion. In this context, the activation of the quadriceps muscles vastus lateralis (VL) and vastus medialis (VM) produce similar knee joint torques. However, VL and VM produce opposite mediolateral patellar forces (internal joint variables), which may result in patellofemoral pain if muscle activations are not properly coordinated. Therefore, in addition to task variables, internal joint variables may be also controlled by the CNS to maintain the integrity of muscles and internal joint structures. In this poster, we will present preliminary results of a study that assessed the hypothesis that the balance of quadriceps muscles activation is altered when mediolateral patellar forces are unbalanced.
In order to apply a controlled mediolateral force on the patella, we implanted bone screws on the patella and the lateral aspect of the femur, and we secured a rubber band (spring) between them. The spring was detached two weeks after the initial implant. Kinematic (joint angles) and spatiotemporal parameters, as well as electromyography (EMG) patterns of VL and VM were analyzed during locomotion of three rats from baseline (before spring attachment) until one week after spring detachment.
In two of the rats, there was a decrease of VL activation after spring attachment, while VM activation maintained similar values. After detachment of the spring in these two rats, VL activation went back to baseline values, with little alteration in VM activation. In the third rat,

VM activation increased in response to spring attachment and went back to baseline values after detachment, while VL activation was similar all the time. These results are consistent with the hypothesis that the CNS regulates internal joint variables, countering the lateral patellar force by altering the balance between VM and VL activation. In addition to these alterations in muscle activations, spatiotemporal and kinematic parameters changed after spring attachment in the three rats. Most of the values went back to baseline values after spring detachment.
We are currently assessing two more rats. If the biomechanical and muscle activation analysis show similar results to those obtained for these three rats, that would strengthen our hypothesis that CNS alters the balance of quadriceps muscles activation when mediolateral patellar forces are unbalanced.

Disclosures: F. Barroso: None. **C. Alessandro:** None. **T. Sandercock:** None. **M. Tresch:** None.

Poster

410. Posture and Gait: Animal Models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 410.03/NN23

Topic: E.06. Posture and Gait

Support: NIH Grant NS086973-01

Title: Adaptation to quadriceps paralysis as a window into neural control of internal joint variables

Authors: *C. ALESSANDRO[1], B. RELLINGER[1], F. BARROSO[1], T. G. SANDERCOCK[1], M. C. TRESCH[2]
[1]Physiol. department, [2]Biomed. Eng, Physical Med. and Rehab, Physiol., Northwestern Univ., Chicago, IL

Abstract: Scientists have typically considered the question of how the central nervous system (CNS) determines motor commands in terms on task performance. In this context, muscle activations should generate joint torques that successfully accomplish the desired behavioral goal.
In our work we recognize that, in addition to task variables, muscle contractions also influence internal joint structures, leading to ligament strains and contact stresses. Poor regulation of these internal joint variables may cause pain and injuries. We therefore hypothesize that the CNS determines motor commands not only based on behavioral constraints, but also based on the need of maintaining joint integrity.
To test this hypothesis, we examine the neural control of quadriceps muscles acting at the rat knee. We show that activation of any quadriceps muscle lead to similar extension torques (task

variables). On the other hand, vastus medialis (VM) and vastus lateralis (VL), produce opposite mediolateral forces on the patella (internal joint variables). This renders the rat knee an ideal model to disambiguate a pure task-variable control, from a strategy that balances the actions of VM and VL to reduce contact stresses within the knee capsule.
In particular, such a conceptual simplicity allows us to formulate clear predictions on how the CNS may adapt to VL paralysis. One possibility may be to only increase the activity of VM. This strategy would be able to restore the lost knee extension torque, but it would produce unbalanced mediolateral forces on the patella. On the other hand, a control strategy that regulates both task and internal joint variables would increase both VM and RF, with a preference on RF, in order to minimize mediolateral patellar movements. Finally, since RF is a multi-articular muscle, a sole increase of RF activity would lead to extra hip flextion torque that would require additional compensation.
We paralyzed VL in a group of rats (n=6) by transecting the corresponding branch of the femoral nerve, and we observed the subsequent adaptation pattern. More than a week after VL paralysis, subjects showed partial recovery of baseline locomotor kinematics (before nerve cut). Their EMG activity patterns exhibited a large increase of RF, and a minimal increase of VM in all animals. These results support our hypothesis that the CNS regulates both task and internal joint variables. Additional experiments will be performed to validate these results, and to investigate the afferent signals that drive this mechanism. Establishing whether the CNS regulates internal joint variables would have impact on our interpretation of neural control of movements both in healthy and pathological subjects.

Disclosures: C. Alessandro: None. **B. Rellinger:** None. **F. Barroso:** None. **T.G. Sandercock:** None. **M.C. Tresch:** None.

Poster

410. Posture and Gait: Animal Models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 410.04/NN24

Topic: E.06. Posture and Gait

Support: NIH grant NS086973

Title: Correlations between quadriceps muscles during locomotion in the rat

Authors: *M. C. TRESCH[1,3], C. ALESSANDRO[1], F. BARROSO[2]
[1]Biomed. Eng, Physical Med. and Rehab, Physiol., [2]Northwestern Univ., Chicago, IL; [3]Shirley Ryan AbilityLabs, Chicago, IL

Abstract: We have previously shown that in rats the quadriceps muscles vastus medialis (VM) and vastus lateralis (VL) produce very similar forces across the skeleton; when each muscle is

stimulated, the forces measured at the distal tibia are nearly identical. On the other hand, stimulation of VM and VL produces opposing mediolateral forces on the patella. As a consequence, unbalanced activation of these muscles would lead to a net mediolateral force on the patella, causing abnormal contact stresses against the patellar groove on the femur and potentially leading to patellofemoral pain or osteoarthritis. Thus, while VM and VL are synergists in terms of their contributions to task performance variables, they are antagonists in terms of their contributions to internal joint stresses.
We are evaluating whether the nervous system considers these contributions to joint variables when choosing muscle coordination patterns during behavior. In this study we consider the step to step correlations between quadriceps muscle activations during locomotion in rats. We hypothesized that if the nervous system regulated the net mediolateral forces on the patella, there should be a strong positive correlation between VM and VL; i.e. on steps where the activation of VM was high, the activation of VL should also be high in order to minimize any potential net mediolateral patellar force. The correlations between VM and VL to rectus femoris (RF) or to vastus intermedius (VI) might be weaker since those muscles should not produce substantial mediolateral patellar forces.
We therefore recorded quadriceps EMG activity during treadmill locomotion in rats. We examined only stance related EMG activity when all muscles were active, calculating the integrated EMG during each step. In 7 animals, we found that VM and VL were in fact strongly correlated while the correlation between these muscles and RF was considerably lower, consistent with the hypothesis that the nervous system balances mediolateral forces on the patella. We have also performed preliminary experiments in 2 animals recording activity from VI, finding a consistent strong correlation to VM, a less consistent correlation to VL, and a consistent weak correlation to RF. Taken together, these results support the hypothesis that the nervous system regulates quadriceps muscle activations in order to minimize mediolateral stresses on the patella. By coordinating VM and VL activations, the nervous system avoids development of abnormal mediolateral patellar forces. The preliminary observation here, that VI appears to be correlated with VM and, to a lesser extent VL, is harder to explain in terms of neural control of mediolateral forces.

Disclosures: M.C. Tresch: None. **C. Alessandro:** None. **F. Barroso:** None.

Poster

410. Posture and Gait: Animal Models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 410.05/NN25

Topic: E.06. Posture and Gait

Title: In pursuit of a generalized model of legged locomotion

Authors: *G. ANTONIAK[1], V. BHANDAWAT[1], T. BISWAS[2]
[1]Duke Univ., Durham, NC; [2]Loyola Univ., New Orleans, LA

Abstract: Legged locomotion is complex. However, center of mass (COM) movement is simpler and can be fit with simple mechanical models. The inverted pendulum (IP) and the spring-loaded inverted pendulum (SLIP) are two such models. While the IP and SLIP models have been successful in explaining the COM trajectory during walking and running, respectively, they have inherent limitations. IP and SLIP can only model the radial forces, which act along the length of the leg. They are not able to model the tangential forces, which act perpendicular to the leg. Thus, IP and SLIP cannot model cases in which the ground reaction force (GRF) does not directly act on the COM. Because of their inability to model the tangential forces, IP and SLIP do not possess a mechanism for modulating stance duration - the time that the leg is in contact with the ground. Their limitations are especially evident when applied to the locomotion of Drosophila melanogaster, which walks at slow size-specific speeds. The gravitational forces left unopposed by IP and SLIP would necessitate that the fly walk faster than is experimentally observed. The angular spring-loaded inverted pendulum (aSLIP) is a model that successfully describes the fly's locomotion. In aSLIP, an angular spring is added to the IP model. This spring resists motion away from the vertical position of the leg, which occurs at midstance. The angular spring serves as a mechanism of modelling tangential forces, which help to overcome the effects of gravity. By changing the stiffness of the spring, the stance duration of the organism can be tuned. The combination of aSLIP and SLIP successfully predicted the COM trajectory of the fly based on experimental data. This generalized SLIP (GSLIP) model is a likely candidate for a general model of locomotion over all speeds, since aSLIP is optimal for slower locomotion, and SLIP for faster locomotion. The generalizability of the model was evaluated by applying it to the locomotion of several animals including humans, where it predicted the characteristic vertical GRF during walking with a minimum occurring at midstance. Neither IP nor SLIP can predict this characteristic vertical GRF without the addition of transmission dynamics.

Disclosures: G. Antoniak: None. **V. Bhandawat:** None. **T. Biswas:** None.

Poster

410. Posture and Gait: Animal Models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 410.06/NN26

Topic: E.06. Posture and Gait

Support: NSERC Discovery Grant RGPIN201603790

Title: The role of ankle extensors when adjusting to treadmill speed in chronic spinal cats

Authors: *J. HARNIE, C. CÔTÉ-SARRAZIN, M.-F. HURTEAU, Y. THIBAUDIER, C. DAMBREVILLE, E. DESROCHERS, A. DOELMAN, T. ROSS, A. TELONIO, A. FRIGON
Physiologie, Univ. De Sherbrooke, Sherbrooke, QC, Canada

Abstract: Modulating locomotor speed is critical for goal-oriented behaviors, requiring temporal and spatial kinematic adjustments in the gait pattern, as well as in the neural drive to muscles. Studies in spinal-transected cats have shown that adjustments to speed can be mediated by spinal locomotor circuits interacting with somatosensory feedback from the hindlimbs. However, how this is accomplished is poorly understood. To investigate the role of ankle extensors in controlling speed, we performed a unilateral denervation of the lateral gastrocnemius and soleus (LGS) muscles in spinal cats. Seven cats were implanted with electrodes to chronically record electromyography (EMG). The spinal cord was then transected at low thoracic levels and cats were trained to recover hindlimb locomotion. After stable hindlimb locomotion recovered, data (EMGs and kinematics) were collected at speeds ranging from 0.1 to 1.0 m/s in 0.1 m/s increments. The LGS nerve was then sectioned unilaterally. Data collection resumed 1-2 days later, up to 8 weeks post-denervation. All cats could step 1-2 days after denervation, albeit with some alterations in the pattern, particularly at slow and fast speeds. In some cats, paw placement was impaired at contact, with the digits curled into flexion. At the slowest speed of 0.1 m/s, the limb ipsilateral to the denervation consistently took two steps for every step of the contralateral limb in 5 cats. This 1:2 coupling between the contralateral and ipsilateral limbs was due to a more rostral position of the paw relative to the hip at the stance-to-swing transition. At the fastest speeds (0.9 m/s and 1.0 m/s), some cats had difficulty maintaining an alternating gait, transitioning to gallop. These alterations in the pattern gradually disappeared over time. The mean EMG amplitude of extensors generally increases linearly with speed. To investigate this modulation, we measured the regression coefficient (R^2) of the ipsilateral medial gastrocnemius (iMG) at 3 time points in 3 cats. The mean amplitude of iMG increased linearly with speed before denervation ($R^2 = 0.94$), whereas this modulation was lost 1-2 days after denervation ($R^2 = 0.01$) before returning 1-8 weeks later ($R^2 = 0.64$). The loss of modulation early after denervation was because of the relatively larger increase in iMG amplitude at slower speeds. The present results demonstrate that ankle extensor muscles play a major role in the control of hindlimb locomotion, particularly at slow and fast speeds, and that spinal circuits interacting with somatosensory feedback are capable of remarkable compensation when modulating locomotor speed.

Disclosures: J. Harnie: None. **C. Côté-Sarrazin:** None. **M. Hurteau:** None. **Y. Thibaudier:** None. **C. Dambreville:** None. **E. Desrochers:** None. **A. Doelman:** None. **T. Ross:** None. **A. Telonio:** None. **A. Frigon:** None.

Poster

410. Posture and Gait: Animal Models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 410.07/NN27

Topic: E.06. Posture and Gait

Support: NSERC Discovery Grant RGPIN201603790

Title: The modulation of forelimb and hindlimb muscle activity with increasing treadmill speed before and after an incomplete spinal cord injury in adult cats

Authors: *A. W. DOELMAN, T. ROSS, M.-F. HURTEAU, E. DESROCHERS, Y. THIBAUDIER, C. DAMBREVILLE, A. TELONIO, A. FRIGON
Physiologie-Pharmacologie, Univ. De Sherbrooke, Sherbrooke, QC, Canada

Abstract: A change in speed is required for goal-oriented behaviors. The CNS modulates speed by controlling spinal motoneuronal outputs, which can be estimated during real movements by recording the electrical activity of muscles (EMG, electromyography). After an incomplete spinal cord injury (SCI), people often walk slower and have difficulty modulating their speed. Despite its importance, the modulation of motor outputs with increasing speed after incomplete SCI is poorly understood. The purpose of the present study was to investigate the modulation of EMG activity as a function of speed before and after an incomplete SCI in adult cats. Eight cats were chronically implanted with EMG electrodes in several fore- and hindlimb muscles. Data (EMGs and kinematics) were recorded during treadmill locomotion at speeds ranging from 0.4 to 1.0 m/s in 0.1 m/s increments. After obtaining data in the intact state, a lateral spinal hemisection was made at mid-thoracic levels on the right side. Recordings resumed 1 week later and up to 8 weeks after hemisection. After hemisection, cats were trained 5 times a week for 15-20 min to recover or maintain an adequate quadrupedal locomotion. Mean EMG amplitude generally increases linearly with speed during locomotion. To determine if this linear modulation was maintained or altered after SCI, we measured the regression coefficients (R^2) of selected muscles in all four limbs before, 1 week and 8 weeks post-hemisection. In hindlimb extensors (vastus lateralis or lateral gastrocnemius) contralateral to the hemisection (left hindlimb), the modulation of mean EMG amplitude was linear in the intact state ($R^2 = 0.92$). This linear modulation was lost 1 week after hemisection ($R^2 = 0.27$) but recovered 8 weeks later ($R^2 = 0.96$). In hindlimb extensors ipsilateral to the hemisection (right hindlimb), the linear modulation in the intact state ($R^2 = 0.96$) was also reduced 1 wk after hemisection ($R^2 = 0.42$) before returning at 8 wks ($R^2 = 0.85$). In forelimb extensors (triceps brachii), the linear modulation with speed was largely maintained with R^2 values ranging between 0.89 to 0.93 and 0.76 to 0.93 for the ipsilateral and contralateral forelimb, respectively. Therefore, these preliminary results suggest that the linear modulation of EMG amplitude in hindlimb extensor muscles is altered after a lateral spinal

hemisection and that it recovers over time, whereas the modulation in forelimb extensors is generally unaffected. Changes in the modulation of EMG amplitude in hindlimb extensors with speed could explain some of the deficits observed after hemisection, as well as provide clues regarding the impairment in modulating walking speed in people with incomplete SCI.

Disclosures: A.W. Doelman: None. **T. Ross:** None. **M. Hurteau:** None. **E. Desrochers:** None. **Y. Thibaudier:** None. **C. Dambreville:** None. **A. Telonio:** None. **A. Frigon:** None.

Poster

410. Posture and Gait: Animal Models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 410.08/NN28

Topic: E.06. Posture and Gait

Support: NSERC Discovery Grant RGPIN-2016-03790

Title: The spinal control of hindlimb muscle synergies during locomotion

Authors: E. DESROCHERS, M.-F. HURTEAU, J. HARNIE, A. DOELMAN, A. TELONIO, *A. FRIGON
Dept. of Pharmacology-Physiology, Univ. de Sherbrooke, Sherbrooke, QC, Canada

Abstract: The mammalian body is a highly complex system composed of a large number of elements (bones, joints, muscles) with multiple degrees of freedom. It has been proposed that the CNS simplifies the control of movement by co-activating groups of muscles with similar actions, termed muscle synergies. However, where in the CNS muscle synergies are controlled is poorly understood. The basic pattern of locomotion is produced by a network of neurons within the spinal cord called the central pattern generator (CPG). To determine if the spinal locomotor CPG coordinates muscle synergies, we obtained the electromyography (EMG) of several hindlimb muscles during locomotion in cats before and after a complete spinal transection. Two cats were trained to step on a treadmill at different speeds and implanted with electrodes for chronic EMG recordings. After obtaining data (EMG, kinematics) in the intact state at speeds ranging from 0.4 to 1.0 m/s, a spinal transection was made at low thoracic levels and cats were trained to recover hindlimb locomotion. Data collection resumed once the cats achieved stable hindlimb locomotion. To characterize muscle muscles synergies, EMG burst onsets and offsets were determined by visual inspection from the raw EMG waveforms using a custom-made program. A cluster analysis was then applied to group muscles based on their activation time (Krouchev et al. 2006 J Neurophysiol, 96: 1991-2010). In the intact state, we identified 5 muscle synergies that included two flexor and three extensor synergies. Flexor synergy 1 included the semitendinosus and semimembranosus, and was related to knee flexion. Flexor synergy 2 included the tensor fasciae latae, anterior sartorius, medial sartorius, tibialis anterior and extensor digitorum longus,

and was related to hip and ankle flexion. Extensor synergy 1 included peroneus longus, soleus and flexor digitorum longus, and was related to ankle extension. Extensor synergy 2 included flexor hallucis longus, medial gastrocnemius and plantaris, and was related to ankle extension. Extensor synergy 3 included vastus lateralis, vastus medialis, anterior biceps femoris and posterior biceps femoris, and was related to knee and hip extension. The same five synergies were present during hindlimb locomotion after spinal transection. Therefore, these preliminary results indicate that hindlimb muscle synergies during locomotion are primarily controlled by the spinal locomotor CPG interacting with somatosensory feedback from the periphery.

Disclosures: E. Desrochers: None. **M. Hurteau:** None. **J. Harnie:** None. **A. Doelman:** None. **A. Telonio:** None. **A. Frigon:** None.

Poster

410. Posture and Gait: Animal Models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 410.09/NN29

Topic: E.06. Posture and Gait

Title: Selective genetic ablation of adrenergic cells leads to dramatic and progressive motor control and metabolic deficits in adult mice

Authors: S. MANJA[1], A. OWJI[1], L. LINDO[1], J. ALTIER[2], K. ROBY[1], R. RASOOL[1], S. NANDINI[1], J. AYALA[4], S. KING[1], *S. N. EBERT[3]
[1]Biomed. Sci., [2]Med., [3]Burnett Sch. of Biomed. Sciences/College of Med., Univ. of Central Florida, Orlando, FL; [4]Sanford Burnham Prebys Med. Discovery Inst., Orlando, FL

Abstract: Phenylethanolamine N-methyltransferase (Pnmt) converts norepinephrine to epinephrine, and thus serves as a marker for adrenergic cells. To determine the roles of adrenergic cells *in vivo*, we created a novel mouse model by crossing *Pnmt-Cre* knock-in mice with *ROSA26-eGFP-DTA* mice resulting in expression of the *Diptheria Toxin A (DTA)* gene exclusively in Pnmt+ cells, leading to selective ablation of these cells due to DTA-induced apoptosis. *Pnmt-Cre/DTA* mice appeared relatively normal and healthy until they reached adult stages, but soon thereafter (2-3 months of age), began to exhibit labored and discoordinated gait movements, diminished grip strength, decreased nocturnal ambulatory activity, and inability to maintain balance on Rotorod and balance beam tests. Further testing revealed that hindlimb reflexes were significantly impaired relative to control mice as early as 1 month after birth. Remarkably, the *Pnmt-Cre/DTA* mice also failed to gain weight beyond 2-3 months of age despite no significant differences in food or water intake relative to age-matched sibling controls. Evaluation of body composition revealed that the lack of weight gain was likely due, in part, to lack of accumulation of fat and, to a lesser extent, lean body mass. None of these phenotypes were observed in control (*Pnmt-Cre* without *DTA*, or *ROSA26-eGFP-DTA* without *Cre*) mice,

nor were they observed in *Pnmt*$^{-/-}$ (knockout) mice lacking the ability to produce epinephrine. These results indicate that the observed motor and metabolic phenotypes are due to selective loss of adrenergic cells. To determine the cellular anatomical correlates of the motor and metabolic phenotypes observed, we are evaluating adrenergic-derived neurons in the cerebellum and hypothalamus, respectively, through immunofluorescent histochemistry and cellular fate-mapping experiments focused in these regions of the brain known to be important motor and metabolic control centers, respectively, where Pnmt expression has previously been observed. These studies offer exciting new and unexpected insights into the roles of adrenergic neurons in the CNS that could potentially lead to novel therapeutic strategies/targets for treating motor control and/or metabolic disorders.

Disclosures: S. Manja: None. **A. Owji:** None. **L. Lindo:** None. **J. Altier:** None. **K. Roby:** None. **R. Rasool:** None. **S. Nandini:** None. **J. Ayala:** None. **S. King:** None. **S.N. Ebert:** None.

Poster

410. Posture and Gait: Animal Models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 410.10/NN30

Topic: E.06. Posture and Gait

Title: Injured corticospinal tracts prevented learning of split wheel walking in mice

Authors: *J. SONSINI[1], A. CHUGHTAI[2], Z. AHMED[1]
[1]Biol., Grad. Ctr. City Univ. of New York, New York, NY; [2]Biol., CUNY Col. of Staten Island, New York, NY

Abstract: Corticospinal tract (CST) damage is a cause of motor impairment in prevalent acquired neurologic conditions such as spinal cord injury and stroke. The CSTs are responsible for distal fine motor control used in precision gait (Lemon, 2008). Functional ambulation includes the ability to step in different directions. When quadrupeds or humans walk on a treadmill (TM) their limbs step in the opposite direction to the belt (Musienko, et al. 2012). In a hybrid condition, in which one belt moves forward and the other backward healthy adults can easily walk with their legs moving in opposite directions (Choi & Bastian, 2007). We sought to determine whether: 1.) mice could learn hybrid walking, 2.) connectivity patterns between motor cortices and sciatic nerves changed over learning 3.) CSTs are necessary for hybrid walking. We analyzed stepping strategies during hybrid walking coded from video using frame by frame analysis. Strategies were categorized into 6 types. Types 0-3 involved turning repeatedly or dragging limbs for 3 seconds or more, types 4-6 involved increasing levels of left/right and fore/hind limb coordination. Control and mice with histologically confirmed bilateral pyridomoty (Bpx) were examined in free and head restrained conditions before, during, and 4 weeks after an intensive training. Training consisted of walking on a custom made motorized

wheel in which one side of the wheel moved forward and the other backward at the same speed for 2 hrs/day for 10 days**.** We also analyzed gait during TM walking before and after hybrid training. To measure connectivity, 4 different structures were reciprocally tested: sensorimotor cortices and sciatic nerves. Control mice were able to learn and retain strategies 5-6 and never used strategies 0-3. Unrestrained control mice learned in 4 days of training; restrained who were prevented from turning or rearing learned within 2 hours. In contrast, all Bpx mice were never able to learn hybrid walking. Bpx animals never demonstrated stepping strategies types 4-6 and were unable to move their limbs opposite to the direction of the wheel. Before and after training, bPX mice performed TM walking with similar limb coordination as control mice but at slower speeds. Electrophysiological results showed changes in reciprocal connectivity between the involved sites. We concluded that intensive training is not sufficient for mice with Bpx to learn a complex locomotion skill such as hybrid walking. These results indicate that the CSTs are important for learning complex locomotion.

Disclosures: J. Sonsini: None. **A. Chughtai:** None. **Z. Ahmed:** None.

Poster

410. Posture and Gait: Animal Models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 410.11/DP10/NN31 (Dynamic Poster)

Topic: E.06. Posture and Gait

Support: ARO - W911NF1410141 - 64929EG

Title: Uncovering the structure of the mouse gait controller with mechanical and neural perturbations

Authors: *A. VAHEDIPOUR[1], P. SHAMBLE[2], O. MAGHSOUDI[1], M. SHORT[1], B. ROBERTSON[1], A. SPENCE[1]
[1]Bioengineering, Temple Univ., Philadelphia, PA; [2]FAS Ctr. for Systems Biol., Harvard Univ., Cambridge, MA

Abstract: Locomotion is essential to survival in most animals. Studies have shown that animals, including humans, choose a gait that minimizes the risk of injury and maximizes energetic efficiency. Individuals often encounter obstacles and perturbations during normal locomotion, from which they have to recover. Despite the importance of understanding the mechanisms that enable recovery from perturbations, ethical and experimental challenges have prevented full exploration of these in legged systems.
A powerful paradigm with which to tackle this difficulty would be the application of external and internal manipulation of the nervous system. These perturbations could target how gait is regulated and how the neural systems process sensory information to control locomotion during

an unexpected perturbation.
The study here presents data on the behavioral and gait response of mice to rapid, precisely timed, and spatially confined mechanical perturbation applied by a treadmill system. Our data elucidate that after the mechanical perturbation, the mouse gait control structure pushes away from the ideal trot to potentially less stable gaits, and mostly towards bounding. We quantified this shift by projecting the observed gait during each stride onto the line between trot and bound, in the space of quadrupedal gaits. We refer to the value of this projection as λ. For $\lambda = 0$, the gait is the ideal trot; for $\lambda = \pi$, it is the ideal bound. We found that the substrate perturbation caused a significant shift in λ towards bound during the stride in which the perturbation occurred (linear mixed effects model: $\Delta\lambda = 0.14 \pm 0.07$; random effect for animal, $p = 0.041$, $n = 8$ mice). We hypothesize that this is because the bounding gait is better suited to rapid acceleration or deceleration; absorbing or injecting kinetic energy. To determine if the gait shift from trot towards bound is a result of a fear response rather than a response to the mechanical stimulus of the substrate perturbation, we will carry out control experiments using a predator odor as a fearful stimulus.
To evaluate whether the same structure of gait controller exists when undergoing an entirely different class of manipulation, we will apply an internal, neuromuscular perturbation. We will directly stimulate the hamstring muscle using implanted electrodes and a custom magnetic headstage. If the same control structure generalizes across external and internal perturbations, then we expect a similar gait structure that prefers bound during recovery.

Disclosures: **A. Vahedipour:** None. **P. Shamble:** None. **O. Maghsoudi:** None. **M. Short:** None. **B. Robertson:** None. **A. Spence:** None.

Poster

410. Posture and Gait: Animal Models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 410.12/NN32

Topic: E.06. Posture and Gait

Support: NASA Training Grant NNX12AN24H

Title: Simulated cockroach walks in different directions at cockroach-like speeds and stepping frequencies

Authors: ***N. S. SZCZECINSKI**[1], S. E. RUBEO[1], R. E. RITZMANN[2], R. D. QUINN[3]
[2]Biol., [3]Mechanical and Aerospace Engin., [1]Case Western Reserve Univ., Cleveland, OH

Abstract: In this work, we refine a previously developed neuromechanical model of cockroach locomotion (Szczecinski et al. 2014, doi: 10.1007/s00422-013-0573-3). Our neural model contains about 1800 dynamical neurons and about 3300 synapses. The mechanical body has 30

actuated degrees of freedom, each with passive and active components. Each degree of freedom is actuated by an antagonistic pair of Hill muscles. Each pair of muscles has stretch and force feedback reflexes, a CPG that produces oscillation, leg position and force feedback pathways for coordinating inter-joint motion, and position and force feedback pathways for coordinated inter-leg stepping (gait). The model walks at cockroach-like speeds (20 cm/s) in a continuum of directions. This continuum is produced by sparse, abstract descending commands that alter motion only by altering leg-local reflexes (Martin et al. 2015, doi: 10.1016/j.cub.2015.09.044). A major challenge in building a useful model is picking parameter values that produce the intended dynamics. This poster presents a rapid method for selecting parameter values for the neurons, synapses and muscles of the model, totaling about 20,000 parameters. Rather than optimizing or evolving the parameter set that produces locomotion, we tune small, functional subnetworks, which when assembled produce the intended walking. Exploiting the structure of the nervous system to tune parameters eliminates dependence on black-box approaches that may capture animal behavior, but provide no insight into how the system functions.

Disclosures: N.S. Szczecinski: None. **S.E. Rubeo:** None. **R.E. Ritzmann:** None. **R.D. Quinn:** None.

Poster

410. Posture and Gait: Animal Models

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 410.13/NN33

Topic: E.06. Posture and Gait

Support: VR Sweden

Title: Hypoxia Ischemia mice show behavior deficiency on DigiGait and Intelicage behavior test

Authors: *C. XIE
CBR, Univ. of Goteborg, Stockholm, Sweden

Abstract: Background: Different tests were used to measure the behavior alternation after Hypoixa Ischemia (HI) on mice, but most of them are not suitable or sensitive enough. We aim to test two new HI behavior tests (DigiGait, Intelicage) and four other widely used behavior tests to find the most suitable tests for HI studies. **Method:** Male and female mice were subjected HI on postnatal 9 days**.** Injury was valued by infarction volume from MAP-2 staining**.** Six different behavior tests were used including DigiGait, Intelicage, Open field, Object recognition, Rotarod, and Elevate plus maze. **Result:** Intellicage showed HI mice performed less total visits and nose pokes and less incorrect visits ration and nose pokes ratio in both male and female mice. DigiGait show left and right fore paw perform differently in male mice. There is No difference between 2 groups in other behavior tests expect Rotarod test show HI mice have significant

lower riding time 31.08s compare with control group 54.92s (p=0.0029). **Conclusion:** Intelicage, DigiGait and Rotarod tests can clearly show difference between HI and control groups, while other tests have no difference. So Intelicage and DigiGait could be used as two new efficient behavior tests together with Rotarod when conducting HI mice studies.

Disclosures: C. Xie: None.

Poster

411. CPGs: Circuit Mechanisms

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 411.01/OO1

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH Grant

ERC Grant

Swedish Research Council Grant

Title: Specificity of excitatory spinal neurons involved in generation of rhythmic activity for locomotion

Authors: *A. E. TALPALAR[1], V. RIBEIRO CALDEIRA[1], T. I. TALPALAR[1], S. M. DYMECKI[2], O. KIEHN[1]
[1]Dept. Neuroscience, Karolinska Institutet, Stockholm, Sweden; [2]Harvard Med. Sch., Boston, MA

Abstract: Locomotion is characterized by repetitive activity of muscles that displace the body in space. The neurons, network structure, and mechanisms that give rise to rhythmic activity in the mammalian motor system are not fully understood. Glutamatergic Shox2- and Hb9-interneurons play an important role in rhythm-generation in mammals (Dougherty et al. 2013; Caldeira et al. 2017). However, experiments in vitro have shown that a minimal inhibitory network can also generate rhythmic activity in presence of drugs and absence of glutamatergic neurotransmission (Talpalar et al 2011). To resolve this apparent discrepancy, we studied the generation of rhythmic activity both in vitro and in vivo using mouse genetics, electrophysiology and behavioral methods. Inactivation of all spinal glutamatergic neurons allowed the production of drug-induced rhythmic-activity in vitro (n=12) but was associated with limb paralysis and lack of postural activity in vivo (n=14), indicating that excitatory neurons are necessary for locomotor function in intact animals. Inactivation of dorsal spinal glutamatergic interneurons produced ataxia and slow locomotion, but did not essentially change other locomotor features (n=7). Inactivation of subpopulations of neurons in particular the Shox2-derived glutamatergic neurons preserved locomotor pattern in vitro and in vivo but reduced its frequency (n= 7). Inactivation of

glutamatergic Hb9-interneurons preserved the production of in vitro rhythmic activity but lead to decline of hindlimb locomotor activity in vivo (n=5). We conclude that while the normal production of locomotor activity in vitro can be replaced by inhibitory networks that are appropriately activated by high concentrations of drugs this is not the case in vivo where spinal glutamatergic neurons are essential for locomotion. Shox2-interneurons are necessary for eliciting high but not low frequency locomotor activity in vivo. While glutamatergic Hb9-interneurons are essential for execution of rhythmic motor activity at all frequencies in in the hindlimbs but not the forelimbs. Together our results confirm that glutamatergic neurons are essential for rhythm generation in the mammalian spinal cord. Supported by NIH, VR and ERC.

Disclosures: **A.E. Talpalar:** None. **V. Ribeiro Caldeira:** None. **T.I. Talpalar:** None. **S.M. Dymecki:** None. **O. Kiehn:** None.

Poster

411. CPGs: Circuit Mechanisms

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 411.02/OO2

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH Grant R01NS090919

NIH Grant R01NS095366

Title: Computational modeling of interactions between cervical and lumbar CPG circuits in the rodent spinal cord *In vitro*

Authors: *N. A. SHEVTSOVA, S. M. DANNER, I. A. RYBAK
Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: The mammalian spinal cord contains central pattern generators (CPGs) that can produce locomotor-like activity in the absence of rhythmic inputs. In quadrupedal animals, each limb is controlled by a separate CPG. The CPGs controlling forelimbs are located in the cervical cord whereas the CPG controlling hindlimbs are located in the lumbar cord. Fictive locomotor activity evoked in the isolated spinal cord of rats by application of neuroactive drugs is characterized by left-right and flexor-extensor alternation in both cervical and lumbar compartments as well as by homolateral alternating and diagonal synchronized activity between the two compartments. Coordination between the left and right circuits in the spinal cord is mediated by excitatory and inhibitory commissural interneurons (CINs). Cervico-lumbar interactions are mediated by ascending and descending ipsi- and contralateral long propriospinal neuron (LPN) connections. If isolated from each other *in vitro* by sucrose blockade, the cervical and lumbar CPGs can independently generate locomotor activity at regular and similar

frequencies (Juvin et al. 2005). Selective activation of the lumbar cord induces coordinated rhythmic activity at the cervical level. In contrast, chemically activating the cervical cord cannot elicit lumbar locomotor activity (Juvin et al. 2005). Midsagittal lesions of the isolated cord at different levels showed that LPN connections contribute to the left-right coordination in fictive locomotion (Juvin et al. 2012; Cowley and Schmidt, 1997). To investigate the role of different LPNs in maintaining the left-right and cervical-lumbar coordination in fictive locomotion we extended our previous models of the spinal locomotor CPGs (Shevtsova et al. 2014; Danner et al. 2016) by incorporating LPNs in accordance with the recent experimental findings (Ruder et al. 2016). The model includes four RGs (left and right cervical and left and right lumbar). Each RG consists of the flexor and extensor half-centers mutually inhibiting each other. Coordination between the left and right RGs is provided by excitatory and inhibitory CINs, including excitatory $V0_V$ and V3 and inhibitory $V0_D$ CINs. Cervical-to-lumbar interactions involve excitatory ascending and excitatory and inhibitory descending LPNs. Our model reproduces the above experimental data and provides explanations for a series of experimental results, including the consequences of selective activation of cervical or lumbar CPGs and midsagittal lesions at different levels. The model provides important insights into the organization of spinal CPG and neural control of locomotion.

Disclosures: **N.A. Shevtsova:** None. **S.M. Danner:** None. **I.A. Rybak:** None.

Poster

411. CPGs: Circuit Mechanisms

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 411.03/OO3

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH Grant R01NS090919

NIH Grant R01NS095366

Title: Computational modeling study of the role of long propriospinal neurons in speed-dependent gait expression

Authors: S. M. DANNER, N. A. SHEVTSOVA, *I. A. RYBAK
Dept. of Neurobio. and Anat., Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: To effectively move in a complex and dynamic environment, limbed animals should vary locomotor speed and adapt gaits to the desired speed. With increasing speed, most quadrupedal animals, including mice, switch from walk to trot and then to gallop and bound. Centrally, the locomotor gaits are controlled by interactions between four central pattern generators (CPGs) located on the left and right sides of the lumbar and cervical enlargements of

the cord, and each producing rhythmic activity controlling one limb. The activity of CPGs are coordinated by commissural interneurons (CINs), projecting across the midline to the contralateral side of the cord, and by long propriospinal neurons (LPNs) that connect the cervical and lumbar CPG circuits in both directions. We use computational modeling to investigate how the LPN connections between the cervical and lumbar CPGs can be organized and what roles different LPN pathways can play in the control and speed-dependent expression of different gaits. Our model contains four rhythm generators (RGs) with left-right cervical and lumbar CIN interactions and homolateral and diagonal ascending and descending LPN interactions. These interactions are organized via several interneuronal pathways mediated by genetically identified neuron types and are based on their suggested functions and connectivity. Supra-spinal (brainstem) drives excite all RGs, thereby controlling oscillation frequency, and inhibit some CINs and LPNs, which allows the model to reproduce the speed-dependent gait transitions observed in the intact mice (Bellardita and Kiehn, 2015). The model reproduces the experimentally observed loss of particular gaits after selective removal of genetically identified neurons (V2a, $V0_V$, or all V0) and the speed-dependent disruption of hind limb coordination after deletion of ascending (cervical-to-lumbar) LPNs (Ruder et al. 2016). The model suggests that (1) $V0_D$ and $V0_V$ CINs together secure left-right alternation, whereas V3 CINs promote left-right synchronization, and that (2) $V0_D$ LPNs support diagonal alternation, whereas $V0_V$ LPNs promote diagonal synchronization. Thus, $V0_D$ CINs and LPNs together stabilize walk and $V0_V$ CINs and LPNs stabilize trot. The transition from trot to gallop and bound occurs when the activity of V3 CINs overcomes the activity of (brainstem-drive inhibited) $V0_V$ CINs and diagonal LPNs. The model proposes a series of testable predictions, including the anticipated effects of the deletion of ascending LPNs, and can be used for simulating various spinal cord perturbations and injuries including hemisections and contusions.

Disclosures: S.M. Danner: None. **N.A. Shevtsova:** None. **I.A. Rybak:** None.

Poster

411. CPGs: Circuit Mechanisms

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 411.04/OO4

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NSERC

Fondation du CHUQ

Title: Genetic dissection of the spinal locomotor circuit in DSCAM mutant mice

Authors: *L. THIRY[1], F. BRETZNER[2]
[1]CRCHUQ, Quebec, QC, Canada; [2]Psychiatrie et Neurosciences, CRCHUQ - Univ. Laval, Quebec, QC, Canada

Abstract: Recently, we have shown that a systemic mutation of DSCAM (Down Syndrome Associated Cell Adherence Molecule) induces anatomical and neurophysiological changes in spinal interneuronal and sensorimotor circuits, contributing to functional locomotor deficits. In order to genetically identify and characterize the neuronal populations underlying these functional changes, DSCAM expression was conditionally impaired in either excitatory or inhibitory neurons by back-crossing VGluT2-cre or vGAT-cre mice with DSCAMf/f mice. During fictive locomotion, neonatal VGluT2-cre;DSCAMf/f mutant spinal cords displayed a left-right synchronization of the ventral root activities, while the flexor-extensor alternation was preserved. Combining retrograde tracing and immunohistochemistry, we found that the number of excitatory spinal commissural interneurons was significantly increased in these neonatal mutant spinal cords. In contrast, the conditional mutation of DSCAM in inhibitory neurons, using VGAT-cre;DSCAMf/f spinal cords, was characterized by an increase in the number of inhibitory spinal commissural interneurons, associated to a normal left-right alternation but a significant reduction in the flexor-extensor coordination during neonatal fictive locomotion. In summary, our studies argue that conditional mutations of DSCAM in either excitatory or inhibitory spinal interneurons can impair different spinal locomotor circuits.

Disclosures: L. Thiry: None. **F. Bretzner:** None.

Poster

411. CPGs: Circuit Mechanisms

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 411.05/OO5

Topic: E.07. Rhythmic Motor Pattern Generation

Support: Swedish Research Council

Swedish Research Council International Postdoctoral Grant

Swedish Brain Foundation

Title: V2a interneuron sub-circuits controlling the speed of locomotion

Authors: I. PALLUCCHI[1], J. SONG[1], K. AMPATZIS[1], J. AUSBORN[2], *A. EL MANIRA[1]
[1]Karolinska Inst., Stockholm, Sweden; [2]Neurobio. & Anat., Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: Animals constantly adjust the speed and coordination of their movements to adapt to changes in the internal or external environment. Yet the circuit mechanisms underlying the change in speed of locomotion remain poorly understood. We have previously shown, in the adult zebrafish, that the spinal locomotor network is not uniform, but consists of three distinct microcircuit modules. These microcircuits comprise three subclasses of the premotor V2a excitatory interneurons which respectively drive the slow, intermediate and fast motor neurons. The speed of locomotion is thus increased through sequential activation of the three modules. However, the logic of V2a interneuron connectivity and how it mediates the precise control of locomotor speed has remained elusive. Here we reveal, using dual patch-clamp recordings in the adult zebrafish, the properties and connectivity pattern of the three V2a interneurons sub-circuits. Each sub-circuit comprises a functionally distinct V2a interneuron subtype forming a reciprocally interconnected excitatory network. Furthermore, V2a interneurons of adjacent sub-circuits display asymmetrical connectivity with higher connectivity probability and synaptic strength from faster to slower sub-circuits (e.g. intermediate to slow and fast to intermediate). A proportion of V2a interneurons within each sub-circuit display intrinsic bursting properties and their minimum bursting frequency directly correlates with their minimum recruitment frequency during locomotion. These intrinsically bursting V2a interneurons are unevenly distributed among the three sub-circuits with a greater number in the slow circuit. The intrinsic bursting properties combined with the connectivity pattern between V2a interneurons within and across sub-circuits may enable the transformation of tonic commands from the brain into rhythmic locomotor activity and increase in speed. Thus, our results reveal an excitatory circuit motif for generation of the locomotor rhythm and smooth change of speed.

Disclosures: I. Pallucchi: None. **J. Song:** None. **K. Ampatzis:** None. **J. Ausborn:** None. **A. El Manira:** None.

Poster

411. CPGs: Circuit Mechanisms

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 411.06/OO6

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NSF Grant IIS-1514622

NIH Grant 1U01EB021921-01

Title: Connectivity analysis of multielectrode neural recordings using a stochastic framework

Authors: *M. ABOLFATH-BEYGI[1], T. D. SANGER[1], S. F. GISZTER[2]
[1]Dept. of Biomed. Engin., USC, Los Angeles, CA; [2]Dept Neurobiol & Anat, Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: We are developing a new methodology for connectivity analysis using a stochastic model along with neural perturbation. The mothod is based on stochastic dynamic operators (SDO) in order to quantify the inter-relations of neural dynamics. Current stimulation techniques for connectivity is based on pairwise neural perturbations. Our goal is to develop a methodology for discovering the connectivity map with the stimulation of much fewer neurons. Since the SDOs quantify uncertainty in the dynamics, we also aim to use this framework for discovering connectivity in the presence of uncertainty in high-level network behavior. The main purposes of developing this framework are detecting neural connectivity and developing predictive models suitable for neuroprostheses. The advantages of SDOs over current neural analysis and predictive models include: 1) its ability to describe state-based dynamic effects of neurons, 2) its methodolgy to embed nonlinear dynamics in a linear frameork, and 3) its capacity to extend pair-wise neural analysis to multi-neural analysis through linear superpositions. SDO is an operator triggered by neuron spikes that maps the probability of current state of movement to the probability of next state. An SDO thus represents both sensory and motor effects in the neuron. We use real neural data collected in spinal frogs, and artificial data from a simulated spinal network as a 'ground truth' network of known connectivity and hierarchy, driven by Hodgkin-Huxley dynamics. This network has been developed by Rybak, Shevtsova, and Markin for spinal cord simulation of populations of pattern generation neurons. The 'fictive' simulated network generates rhythms and motor pool spiking which can be integrated to simulate electromyographic (EMG) recordings. Previous tests on the simulated network validated the use of SDO-based predictive models of EMG as well as interneuron firing activities. Recently, our SDO-based technique for discovering connectivity have been tested on the simulated network. By periodic pulse perturbation of a single population, we are able to find the effective SDO of the perturbed population towards a target population for example a motorneuron pool. With the same data, we quantify the change in the firing patterns of all the other populations caused by perturbation as well as their SDOs towards the target population. Evaluation of these SDOs and the firing activity patterns showed that we are able to discover most of the neural pathways that connect the stimulated population to the target population. Therefore we conclude that the SDOs provide a promising mathematical model for understanding neural connectivity.

Disclosures: **M. Abolfath-Beygi:** None. **T.D. Sanger:** None. **S.F. Giszter:** None.

Poster

411. CPGs: Circuit Mechanisms

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 411.07/OO7

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH R01 NS095366

Wings for Life

Title: Interconnectivity within a spinal locomotor rhythm generating interneurons in mouse

Authors: *N. HA[1], L. YAO[2], K. DOUGHERTY[1]
[1]Drexel Univ. Col. of Med., Philadelphia, PA; [2]Drexel Univ. Col. of Med., philadelphia, PA

Abstract: The central pattern generator (CPG) controlling hindlimb locomotion is located in the thoracolumbar spinal cord. Within the CPG, rhythm generating (RG) interneurons (INs) are responsible for the timing of the movements, providing the rhythmic drive to other downstream neurons in order to generate coordinated locomotor behavior. Recently, a class of INs that express the transcription factor Shox2 has been demonstrated to make up part of the rhythm generating kernel, along with other yet to be identified INs. Given the important function of RG Shox2 INs, detailed analysis of cellular and network properties can provide information about rhythmogenic mechanisms. Here, in order to investigate the interconnectivity between Shox2 INs, we performed whole cell paired recordings from identified Shox2 INs in dorsal horn removed preparations or spinal slices from Shox2::Cre ; Ai9 neonatal (P0-P5) mice. We found that both unidirectional and bidirectional synaptic coupling were present within subsets of the Shox2 IN population. The synaptic delay and shape of the excitatory postsynaptic potentials in unidirectional pairs of Shox2 INs suggested chemical synaptic transmission. Bidirectional coupling, however, was detected at a significantly higher rate. In bidirectional pairs of Shox2 INs, a gap junction blocker, but not glutamatergic receptor antagonists, efficiently abolished the connections, demonstrating exclusively electrical synapses. Similar to electrical synapses demonstrated in other systems, those between Shox2 INs acted as low-pass filters. Further, we examined the interconnectivity within the Shox2 IN population in more mature spinal networks, as electrical coupling may only be present at early postnatal stages. In juvenile mice (P13-P17), we found that electrical coupling persisted between Shox2 INs. Measured coupling coefficients, amplitudes of post-junctional current, and latency of post-junctional current delays were similar between neonate and juvenile age groups, indicating that the strength of electrical transmission did not decline. Experiments in the lab probing interconnectivity between Shox2 INs in slices from adolescent (P23-P29) and adult (>P60) mice are ongoing. Overall, our data demonstrate the presence of chemical transmission and electrical transmission within a subset of Shox2 INs with the latter detected at a higher incidence. We propose that electrical transmission is implicated as one of the mechanisms to promote synchronization of RG Shox2 INs.

Disclosures: **N. Ha:** None. **L. Yao:** None. **K. Dougherty:** None.

Poster

411. CPGs: Circuit Mechanisms

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 411.08/OO8

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NSF-IOS-1455527

Title: Distinct neural mechanisms underlie half center oscillators for homologous swimming behaviors of two sea slug species

Authors: *A. SAKURAI[1], P. S. KATZ[1,2]
[1]Neurosci. Inst., Georgia State Univ., Atlanta, GA; [2]Dept. of Biol., Univ. of Massachusetts, Amherst, MA

Abstract: Rhythmic motor pattern generation often involves neurons with reciprocal inhibition forming a half-center oscillator (HCO). The central pattern generators (CPGs) for swimming in the nudibranch molluscs, *Dendronotus iris* and *Melibe leonina* are both organized as HCOs. Although a phylogenetic analysis indicates that the swimming behaviors are homologous, the synaptic organization of the CPGs are very different. In *Dendronotus*, the swim CPG consists of two pairs of reciprocally inhibitory neurons, Si2 and Si3. Si3 drives the contralateral Si2 through an excitatory synapse and electrical connection, causing them to burst nearly synchronously and in alternation with their contralateral counterparts. Here, we found that Si3 exhibited a hyperpolarization-activated slow inward current. We also found that Si2 had a recurrent inhibitory synapse to the contralateral Si3, which caused the Si3 spike rate to decrease during each burst of the swim motor pattern. Concurrently, the contralateral Si3 gradually depolarized because of the slow inward current activation. The transition of excitation occurred when Si3 escaped from the inhibition of the contralateral side.
In contrast, the *Melibe* swim CPG consists of four bilaterally symmetric swim interneurons (Si1-4), each forming reciprocal inhibition with its contralateral counterpart. The ipsilateral Si1, Si2, and contralateral Si4 are electrically coupled and together form the primary kernel of the HCO. We found no active membrane properties in these neurons that could provide the dynamics needed for rhythmic transition of excitation from one side to the other in the HCO. Instead, the transition was mediated by complex synaptic interactions between the primary kernel and Si3. During the swim motor pattern, a burst in neurons on one side of the primary kernel induced a burst in the contralateral Si3, which in return terminated the burst in the primary kernel. Cessation of bursting on one side of the kernel released the other half to fire a burst, which in turn terminated the ongoing Si3 burst. Thus, the two halves of the primary kernel and each Si3 generated rhythmic activity through a mechanism of recurrent cyclic inhibition. These results demonstrate that although the swimming behaviors are homologous and resemble each other, the neural mechanisms underlying the generation of bursting differ substantially.

Disclosures: A. Sakurai: None. **P.S. Katz:** None.

Poster

411. CPGs: Circuit Mechanisms

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 411.09/OO9

Topic: E.07. Rhythmic Motor Pattern Generation

Title: Multi-compartmental cardiac ganglion large cell model to study single cell and network outputs

Authors: *S. S. NAIR[1], D. C. WOOD[2], J. WANG[2], D. KICK[3], D. SCHULZ[3]
[1]Electrical & Computer Engin., Univ. of Missouri Columbia, Columbia, MO; [2]Biol. Engin., [3]Div. of Biol. Sci., Univ. of Missouri, Columbia, MO

Abstract: The crustacean cardiac ganglion (CG) network in *Cancer borealis* consists coordinates the rhythmic bursting of the heart muscle contractions to control the circulation of blood. Although the network consists only 9 cells, 5 ‘large cell’ motor neurons (LCs) and 4 small endogenous pacemaker cells (SCs), the role of the large cells with variable sets of conductances, in coordinating the features of the network rhythm remain unclear. We developed an improved multi-compartmental biophysical model for the LCs that better represents the morphology including locations of SC-LC synapses. The nominal model used reported conductance data including from our own current measurements. Earlier, we reported single cell model predictions about correlations among conductances that preserved LC response characteristics. The revised LC model is used to first investigate potential distributions of conductances in multiple compartments to preserve the same passive and current injections properties. The set of single LC models from such a study will then enable an investigation of correlations between maximal current conductances, both within a single compartment and across compartments that preserve response characteristics at the single LC level. These LC models with variable underlying conductances are then used in network models to study the effect of variations in the underlying conductances, and of the LC-SC feedback, on the features of the network rhythm.

Disclosures: S.S. Nair: None. **D.C. Wood:** None. **J. Wang:** None. **D. Kick:** None. **D. Schulz:** None.

Poster

411. CPGs: Circuit Mechanisms

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 411.10/OO10

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH grants HD325

NIH grants EB012855

Title: Balancing inhibitory and excitatory coupling in a multifunctional central pattern generator with slow and fast rhythms

Authors: J. R. P. GREEN[1], R. KHWAJA[2], A. N. KLISHKO[3], B. I. PRILUTSKY[4], *G. S. CYMBALYUK[2]
[1]Neurosci., [2]The Neurosci. Inst., Georgia State Univ., Atlanta, GA; [3]Sch. of Applied Physiology, Ctr. for Human Movement Studies, Georgia Inst. of Technol., Atlanta, GA; [4]Georgia Inst. Technol., Atlanta, GA

Abstract: We have investigated whether mammalian central pattern generators (CPGs) could be multifunctional, controlling more than one functional rhythmic behavior. We constructed a model of a single CPG that controls both locomotor-like rhythm (1 Hz) and paw shake-like rhythm (10 Hz) in cats to propose general biophysical mechanisms, supporting vastly different rhythms of a multifunctional CPG. This model consists of four neurons: two inhibitory neurons carry reciprocal inhibition between two excitatory neurons. Each of these neurons represents a neuronal population in the cat CPG. The excitatory neurotransmission is conducted by NMDA and AMPA glutamatergic synaptic currents and the inhibitory neurotransmission is GABAergic and glycinergic. This model exhibits coexistence of the locomotor-like rhythm and the paw shake-like rhythm. The paw shake-like response can be elicited by applying an excitatory pulse of current in either the multistable model or a monostable model. The described mechanism of multistability provides various testable predictions about bursting dynamics throughout a paw shake-like response. These predictions were also tested in a corresponding population model, consisting of two inhibitory populations of 10 neurons each that carry reciprocal inhibition between two excitatory populations of 10 neurons each. The population model also exhibits coexistence of locomotor-like and paw shake-like rhythms and provides predictions consistent with the four neuron model. Our models predict that the flexor burst duration and the extensor interburst interval increase throughout a single paw shake-like response. Adult cats where trained to walk on a walkway using food reward. Hindlimb kinematics and muscle activity were recorded during walking and during paw shake responses; the latter were elicited in cats by attaching an adhesive tape on the hindpaw and letting the cat walk on a walkway. The cats performed paw-shake responses intermittently while walking, and each paw-shake response consisted of 4 to 10 cycles. We found that experimental results were consistent with model predictions. There was a progressive increase in EMG burst duration in consecutive paw-shake cycles for flexors and a progressive increase in EMG interburst interval for extensors. We concluded that a paw-shake response might be a transient response to sensory input to the locomotor CPG.

Disclosures: J.R.P. Green: None. **R. Khwaja:** None. **A.N. Klishko:** None. **B.I. Prilutsky:** None. **G.S. Cymbalyuk:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.01/OO11

Topic: E.10. Motor Neurons and Muscle

Support: European Research Council Consolidator Grant – DBS Model 646923

Title: Factors influencing beta-band motor unit coherence with changining force level

Authors: *L. M. MCMANUS, M. LOWERY
Univ. Col. Dublin, Dublin, Ireland

Abstract: Communication within the neural network may be reflected in the synchronization observed between spatially distinct neural oscillators. The synchronous discharge of motor units, particularly in the beta-band (15-30 Hz) frequency range, has been linked to oscillatory cortical and sub-cortical processes. A reduction in corticomuscular coherence and in synchronization between pairs of motor unit (MU) has been shown with increasing force [1,2]. We examined whether a systematic change occurred in the beta-band coherence among populations of MUs with changing muscle force in the first dorsal interosseous (FDI), and used a model of the motoneuron pool to explore the factors that could alter MU coherence [3].
Beta-band MU coherence was investigated in the FDI (N=6) where subjects increased their force from 20% to 35% MVC, or decreased it from 35% to 20% MVC, midway through a contraction (45s). Each subject performed both the increasing and decreasing force trial. Coherence in the was also investigated in the FDI muscle during separate contractions at 10% and 20%. Discriminable MUs were extracted from the surface EMG using decomposition algorithms (Delsys, Inc.).
In the trials with two force levels (20% and 35%MVC), pooled MU coherence in the beta-band decreased at the higher force level (-75±18%) and increased at the lower force level in all subjects (278±225%). Beta-band coherence was higher in contractions at 10%MVC (19±4) when compared to those at 20%MVC (13±7, $p<.05$). Model simulations suggest changes in both the proportion and strength of common inputs and in motoneuron firing rates could alter the resulting expression of synchronization and coherence among MU firing times.
In this study, beta-band intra-muscular coherence decreased at higher force levels. Several factors may be contributing to the observed reduction. Reduced synchronization of pre-synaptic inputs to the motoneuron pool, slight changes in the frequency of these inputs or in motoneuron firing rates could all alter the resulting expression of synchronization and coherence among MU firing times. In addition, it is possible that the relative strength of common synaptic inputs is weakened in the presence of higher excitatory drive to the motoneuron pool. A parallel increase in synaptic noise could also potentially decrease the synchronization between MU firings without

any change in the proportion of common input to the motor neuron pool.
[1] Ushiyama et al. (2011). J Neurophysiol,106(3),1379-1388.
[2] Kline & DeLuca (2016). J Neurophysiol,115(1),178-192.
[3] Lowery & Erim (2005). J Comp Neurosci,19(2),107-124.

Disclosures: L.M. McManus: None. **M. Lowery:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.02/OO12

Topic: E.10. Motor Neurons and Muscle

Support: De Luca Foundation

NIH Grant R44NS077526

NIH Grant R43NS093651

Title: Common drive of motor units during dynamic activities of the upper-limb

Authors: *J. C. KLINE, P. CONTESSA, S. H. ROY, G. DE LUCA
Delsys, Inc, Natick, MA

Abstract: Studies of motor unit firing behavior have reported varying degrees of “common drive”, or correlated firing rates, depending on whether muscles are activated in synergist or antagonist contractions [1]. Although such studies of isometric muscle activity inform the control mechanisms that govern stationary contractions, there remains a need to determine how the central nervous systems regulates functional tasks of normal voluntary movement. Therefore, we employed recently developed surface electromyographic (sEMG) decomposition technology to investigate the activation of motor units during cyclic movements of the upper-limb. Non-invasive sensors recorded sEMG signals from extrinsic muscles of the hand and forearm during voluntary dynamic activities of opening and closing the hand or grasping different objects. The recorded sEMG signals were processed by algorithms to extract the action potentials and corresponding firing times of the active motor units [2]. The measured motor unit data were validated for accuracy and compared with the force, position and angular velocity of the fingers, hand and arm using the Trigno™ sensor system (Delsys, Inc. Natick, MA). Based on these data we observed that motor unit firing rates within each muscle: 1) were ordered in an inverse hierarchical relationship relative to the motor unit action potential amplitude as previously described by the “onion-skin” phenomenon; and 2) were correlated with respect one-another and with the output movement in accordance with the common drive [2]. When comparing these data across muscles we found that the firing rates of motor units from different muscles maintained a

relatively high degree of correlation across all activities. However, the latency of the firing rate correlation shifted depending on the activity being performed. When two muscles were coactivated as synergists, such as when grasping an object, the correlation of the motor unit firing rates manifested approximately zero latency. Yet when the same muscles were activated as antagonists the degree of correlation remained relatively high but the latency of the correlation increased. These data indicate that the central nervous system coordinates the synergist and antagonist activation of groups of muscles by regulating the latency of the common drive between the motor unit firing rates during different voluntary movements. This gives evidence to the importance of the common drive control scheme for regulating functional voluntary movement. References 1. **De Luca CJ, et al**. *J. Neurophys*. 87:2200-2204, 2001. 2. **De Luca CJ, et al**. *J. Neurophys*. 113:1941-1951, 2015.

Disclosures: J.C. Kline: None. **P. Contessa:** None. **S.H. Roy:** None. **G. De Luca:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.03/OO13

Topic: E.10. Motor Neurons and Muscle

Support: De Luca Foundation

NIH Grant R44NS077526

NIH Grant R43NS093651

Title: Influence of synergist muscle activation on motor unit firing behavior during fatigue

Authors: J. LETIZI, J. C. KLINE, *P. CONTESSA
Delsys Inc, Natick, MA

Abstract: Previous investigations have indicated that the neuromuscular system adapts by concurrently increasing motor unit firing rates and recruitment to sustain force production during fatigue [1]. This control scheme has been verified in the vastus lateralis muscle of the leg [1-2], but it has never been observed in the first dorsal interosseous (FDI) muscle of the hand, which is typically associated with decreasing motor unit firing rates with fatigue [3-4]. These contrasting reports raise questions as to whether the control scheme governing motor unit behavior during fatigue is altered across muscles, or whether other factors such as force compensation from synergist muscles may account for such differences. Therefore, we set out to investigate motor unit firing behavior during a fatigue protocol in the FDI muscle. Seven healthy (22-34 years old) subjects performed voluntary isometric contractions of the FDI sustained for 10 s at 50% maximal voluntary contraction force and repeated to the endurance limit. Motor unit action

potentials and firing times were extracted using surface electromyographic (sEMG) decomposition technology [5]. To monitor the contribution of synergist muscles to index finger abduction force we recorded sEMG signals from the Flexor Carpi Radialis, Extensor Carpi Radialis, Pronator Teres, and Flexor Carpi Ulnaris muscles. As the FDI contractions were repeated to the endurance limit, we found that motor unit firing rate increased while motor unit recruitment threshold decreased. The increasing trend in firing rate stopped or reversed in four subjects. When the increasing trend stopped or reversed, the sEMG root-mean-square (RMS) from the FDI muscle decreased while that of synergist muscles concurrently increased. These data indicate that increased activation of synergist muscles compensates for decreased activation of the FDI muscle. Our findings provide evidence that fatigue-induced adaptations in motor unit firing behavior in the FDI result from increased excitation to the motoneuron pool that is suggested to compensate for decreases in muscle force-twitch that are reported to occur during fatigue [6]. This control scheme is consistent with our previous observations [1]. Contrasting reports of decreasing or stagnant motor unit firing rates during fatigue may not indicate alterations in the control scheme, but may result from changes in the relative contribution of synergist muscles to the output force.

[1] Contessa et al. J Neurophysiol 2016
[2] DeRuiter et al. Eur J Appl Physiol 2005
[3] McManus et al. J Neurophysiol 2015
[4] Enoka et al. J Neurophysiol 1989
[5] Nawab et al. Clin Neurophysiol 2010
[6] Adam and De Luca. J Appl Physiol 2005

Disclosures: **J. Letizi:** A. Employment/Salary (full or part-time):; Delsys Inc. **J.C. Kline:** A. Employment/Salary (full or part-time):; Delsys Inc. **P. Contessa:** A. Employment/Salary (full or part-time):; Delsys Inc..

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.04/OO14

Topic: E.10. Motor Neurons and Muscle

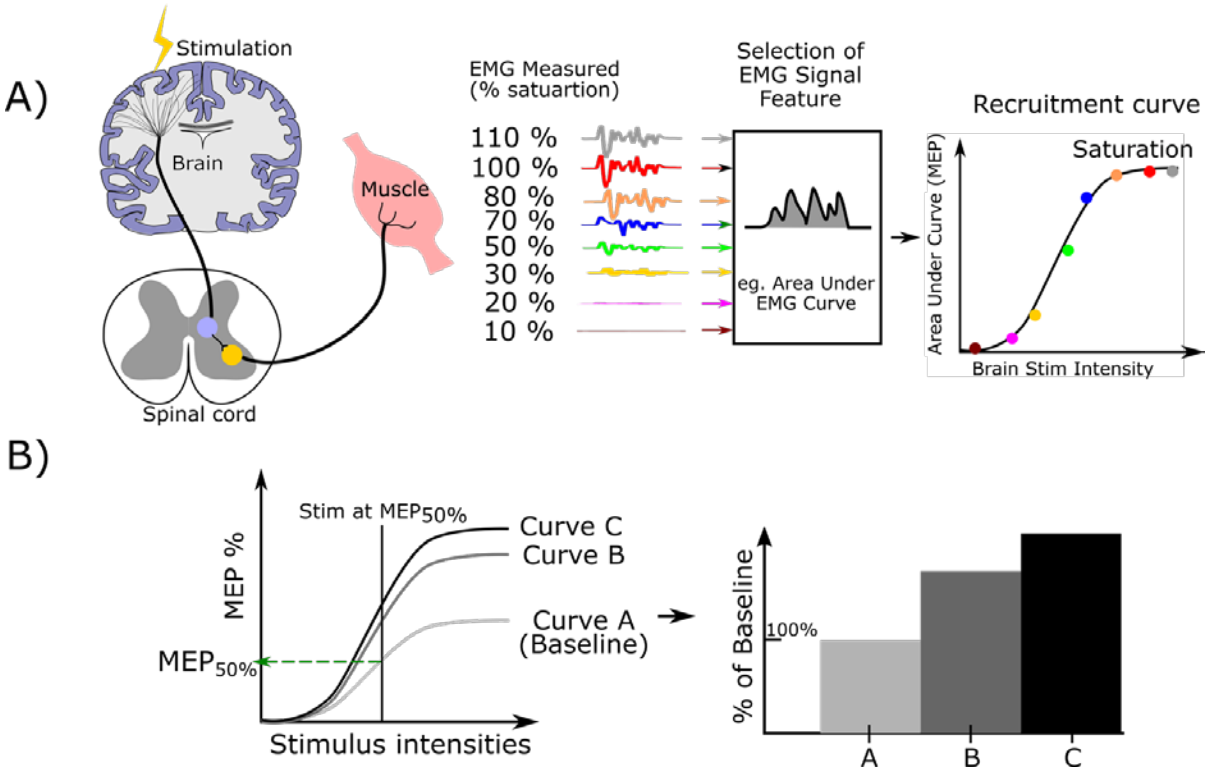
Support: NINDS-NIH 1R01NS092875

Title: Motometrics: A guided analysis toolbox for annotating and analyzing motor evoked potentials

Authors: *S. RATNADURAI GIRIDHARAN[1], D. GUPTA[1,2], J. N. HILL[1,2,3], A. PAL[1], A. M. MISHRA[1], J. B. CARMEL[1,2,3]

[1]Burke Med. Res. Inst., White Plains, NY; [2]Weill Cornell Med., New York, NY; [3]Blythedale Children's Hosp., Valhalla, NY

Abstract: To assay the physiology of the motor system, often the brain is stimulated and the muscle response, or motor evoked potential (MEP), is recorded. To understand the relationship between stimulation intensity and MEP, a recruitment curve is often produced (Fig. 1A). This S-shape curve details the complex input-output relationship between brain stimulation and MEP. Often, one would like to compare multiple recruitment curves that may correspond to different subjects, experimental conditions, and/or different points in time. Generating these curves from data and subsequently analyzing them can be arduous. For this purpose, we provide a software solution called Motometrics. The software uses an intuitive graphical user interface first to annotate and load acquired MEP data independent of the electrophysiological recording system. The user then chooses a signal metric such as the area under the curve to quantify MEPs. The data are fitted to an S-shaped recruitment curve, which in turn can be quantified using several metrics; users can choose a percentage of either the stimulus intensities or MEPs. The metric is applied to all recruitment curves to compare them as shown in Fig 1B. Motometrics is generalizable across humans and animals. It allows the experimenter to perform near real-time analysis of recruitment curves. As an open-source tool (https://bitbucket.org/burkemedicalresearch/motometrics), Motometrics is intended to grow with the needs of the motor physiology community.



Disclosures: **S. Ratnadurai Giridharan:** None. **D. Gupta:** None. **J.N. Hill:** None. **A. Pal:** None. **A.M. Mishra:** None. **J.B. Carmel:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.05/OO15

Topic: E.10. Motor Neurons and Muscle

Title: Motor unit rate coding and recruitment of elbow flexion agonists does not always reflect muscle length-tension curve relationships

Authors: *Z. ADAMS[1], R. E. AKINS, Jr[3], T. S. BUCHANAN[2,1]
[2]Delaware Rehabil. Inst., [1]Univ. of Delaware, Newark, DE; [3]Nemours - Alfred I. Dupont Hosp For Children, Wilmington, DE

Abstract: Muscles produce force via a length-tension relationship, where the force a muscle produces is a function of the number of overlapping fibers during contraction. While relationships between resultant joint kinetics and muscle length-tension curves are well established, little is known about motor unit (MU) activation and muscle length-tension. We hypothesized that MU recruitment and rate coding between elbow flexion agonists would follow normal length-tension relationships. Six subjects (21-29 years) completed a force matching isometric elbow flexion task. Surface electromyography (sEMG) was collected from the right biceps (BB) and brachioradialis (BRA). This task was repeated at various joint angles: 60, 90 and 120 degrees. The task was a ramp to target torque and maintaining for ten seconds. Each task was performed at 30% of the subject's maximal torque. The sEMG was decomposed into individual MU action potential trains. MU train characteristics were obtained from these trains and used for analysis.
BB and BRA MUs surprisingly activated differently with respect to joint angles. A decrease in MU activation was expected as a muscle moves to a more optimal length while maintaining a constant force. BB MU peak firing rates decreased as the muscle length increased ($p<0.05$). Unexpectedly no relationship was found between BRA MU any firing behavior and joint angle. Several factors may play a role in why this occurs. According to normal length-tension curves in the measured range both muscles have an increasing fiber length with an increase in joint angle. This indicates more force production at larger joint angles due to greater fiber overlap. This is demonstrated in the BB data with a decrease in firing rates while maintaining the same resultant torque (30%) without substantial changes to the mechanical advantage of the muscle action. Unlike in the BB the BRA in the ranges measured is closer to optimal fiber length, thus it may not require MU firing changes to maintain a similar load. In addition to muscle length properties BB and BRA also recruited their MUs at different times during force production. BB MUs recruited earlier during the contraction, with BRA recruiting on average 1 second later ($p<0.05$). This is significant when the recruitment ramp was three seconds long. It may be that by recruiting motor units later in the contraction the BRA contributes less to the total joint torque

than the BB, requiring less adjustment along the length-tension curve. These results demonstrate that while BB MUs follow normal muscle length-tension trends the BRA recruits and produces force differently during isometric elbow flexion.

Disclosures: Z. Adams: None. **R.E. Akins:** A. Employment/Salary (full or part-time):; Nemours-A.I. DuPont Research Hospital for Children. **T.S. Buchanan:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.06/OO16

Topic: E.10. Motor Neurons and Muscle

Title: Is increased activation of PFC with cognitive demands associated with the motor unit variability?

Authors: *B. NOH[1], K. C. PHILLIPS[2], H. MAAT[2], T. YOON[3]
[1]KIP, Michgan Tech., Hancock, MI; [2]Kinesiology and Integrative Physiol., Michigan Technological Univ., Houghton, MI; [3]Michigan Tech. Univ., Houghton, MI

Abstract: Exposure to a cognitive challenge impairs motor control by increasing motor unit discharge rate variability and increases activation in the prefrontal cortex (PFC) during low-force contractions. The purpose of this pilot study is to examine whether the increase activation in the PFC is associated with the motor unit variability. Two healthy young men (31.0 ± 5.7 years) participated in this pilot study. A submaximal isometric contraction of the elbow flexor muscles was repeated at 10, 30, 40 and 50% of maximum voluntary contraction (MVC) with and without a mental math task (serial subtractions by 13 from 4-digit numbers). The order of the control (motor task only) and combined task (motor + mental math) were randomized, and the order of the contractions was randomized for each participant but the same throughout each task. Motor control was quantified as force fluctuations (coefficient of variation, CV=100 × Standard deviation of force/mean force). Brain activation was assessed as the change in average oxygenation level (HbO2-HbR) using near infrared spectroscopy (NIRS) over the PFC. Motor unit variability was quantified as discharge rate variability (CV of interspike interval, ISI) of motor units decomposed from surface electromyography signal. Force fluctuations were greater during the combined task compared to the control task at all intensities (2.80 ± 0.39 vs. 4.49 ± 1.43%, $P < 0.05$; averaged value of each intensity). Oxygenation level was increased from baseline ($P < 0.05$) and the increase was greater during the combined task compared to the control task at all intensity (-0.24 ± 0.13 vs. 2.01 ± 0.20 μM, $P < 0.05$). 18-58 motor units decomposed at each contraction were compared, and ISIs were similar between both tasks (0.34 ± 0.01 vs. 0.35 ± 0.00%, $P > 0.05$). No significant correlation was found between oxygenation level with ISI ($r = -0.16$, $P > 0.05$). Increased activation of the PFC was not associated with the

changes in motor unit variability when the cognitive demand was imposed. This result may be due to the small sample size of this pilot study. Further study with large sample size is warranted.

Disclosures: **B. Noh:** None. **K.C. Phillips:** None. **H. Maat:** None. **T. Yoon:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.07/OO17

Topic: E.10. Motor Neurons and Muscle

Support: NSF Grant DGE-11-44155

NIH, NINDS Grant DP2 MH100016-01

Simons Foundation

McKnight Scholar Award

Klingenstein-Simons Fellowship

Searle Scholars Program

Kavli Foundation

Title: Optimal force production by an idealized motor pool necessitates a flexible interplay between size and speed principles

Authors: *N. J. MARSHALL, L. ABBOTT, M. M. CHURCHLAND
Neurosci., Columbia Univ., New York, NY

Abstract: A motor pool consists of ~10^2-10^3 motor units (MUs), providing innumerable ways to generate a desired force. During slow-ramp and steady-state isometric force production, MUs are recruited in ascending rank order by size. At apparent odds with this size principle, there exists evidence for the selective recruitment of large MUs when force must be generated quickly, including during rapid dorsiflexion in humans and running in rats. As larger MUs generally contain faster-contracting muscle fibers, these findings suggest that speed, as well as size, may influence MU recruitment. It seems intuitively appropriate to recruit faster fibers for faster movements, but does this in fact confer advantages?
We explored this question at the theoretical level by computing the optimal firing rates of a pool of five idealized MUs whose twitch responses displayed inversely related peak tensions and contraction times (i.e., larger units were faster twitch). The force contributed by each MU was modeled as the convolution of its twitch response with its spikes, produced via an

inhomogeneous Poisson process. The model's goal was to match the total force to a variety of time-varying target forces: ramps, steps, and sinusoids. Error could arise from two sources: trial-to-trial variability in output force and systematic discrepancies between the average output and target forces. We incorporated both sources of error into a cost functional over the set of firing rates. With the added requirement of non-negative solutions, we formulated the cost functional as a bound-constrained quadratic programming problem and numerically derived the optimal set of MU responses.
For steady-state and linearly increasing forces, optimization yielded the usual size principle: MUs were recruited in ascending order by their peak tension (i.e., slower/smaller units were recruited first). Similarly, units were de-recruited from largest to smallest for slowly decreasing ramp forces. More complex force profiles yielded more intricate optimal strategies. For example, when force had to be quickly decreased, optimal force production required de-recruiting smaller/slower motor units early and compensating with larger/faster motor units. This strategy is optimal because just prior to force offset, most force is generated by faster units whose force output can be rapidly terminated. Similarly complex optimal strategies were seen for sinusoidal forces. These results indicate that optimal force production requires flexible recruitment strategies; the size principle is optimal under only some circumstances. Whether the nervous system displays such flexibility remains an open question.

Disclosures: **N.J. Marshall:** None. **L. Abbott:** None. **M.M. Churchland:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.08/OO18

Topic: E.10. Motor Neurons and Muscle

Support: ERC Consolidator Grant - DBSModel 646923

Title: Evidence of increased synchrony within and between muscles during isometric leg extension in Parkinson's disease

Authors: *M. W. FLOOD[1,3], B. R. JENSEN[4,5], A. MALLING[4,5], M. M. LOWERY[2,3]
[2]Sch. of Electrical & Electronic Engin., [1]Univ. Col. Dublin, Dublin, Ireland; [3]Insight Ctr. for Data Analytics, UCD, Dublin, Ireland; [4]Dept. of Neurology, Odense Univ. Hosp., Univ. of Southern Denmark, Odense, Denmark; [5]Integrated Physiology, Dept. of Nutrition, Exercise & Sports, Univ. of Copenhagen, Copenhagen, Denmark

Abstract: The result of a depletion of dopaminergic neurons of the substantia nigra, Parkinson's disease (PD) is characterised by the triad of motor symptoms - tremor, rigidity and bradykinesia. While Parkinsonian tremor has long been associated with oscillations at frequencies <10 Hz,

enhanced beta band oscillatory activity (12-30 Hz) within the cortico-basal ganglia network has more recently been linked to bradykinesia and rigidity in individuals with PD. To examine whether evidence of increased neural synchrony is present in the motor neuron pool in PD, coherence and nonlinear signal processing methods were used to investigate differences in EMG signal structure in simultaneously active muscles during isometric leg extension. These methods were also applied to simulated EMG data to assess the influence of enhanced synchronous oscillations to the motor neuron pool on the structure of the surface EMG signal.Surface EMG was recorded from extensor and flexor muscles of the upper leg in 13 controls (65 ± 6yrs.) and 13 PD patients (63 ± 6yrs.) during isometric leg extension at 15% of maximum voluntary contraction. Intermuscular coherence was estimated using 60s rectified EMG signals concatenated from 15s sections from each trial. Determinism (%DET) and sample entropy (SampEn) were estimated from 9 overlapping windows (1.5s) in each trial. Surface EMG was simulated *a posteriori* using a model of EMG from two agonist leg muscles with an additional common modulation current (17.5-22.5 Hz) to the motoneuron pool.Average intermuscular coherence between agonist muscle pairs was significantly higher in PD than controls in the alpha (pre: $p = 0.014$, post: $p = 0.029$) and beta (pre: $p = 0.013$, post: $p = 0.011$) bands. %DET was higher and SampEn was significantly lower in the PD group. Intermuscular coherence of simulated EMG increased in the presence of a common beta modulation to the motoneuron pool. SampEn was lower and %DET higher in the simulated EMG in the presence of common input. Values were similar to those obtained from experimentally recorded EMG, when the common modulation accounted for 5% of the total current input to each motoneuron.
This study identified differences in intermuscular coherence between individuals with PD and age-matched controls during an isometric leg extension. The higher levels of intermuscular coherence and reduced complexity observed in EMG of individuals with PD, and in simulated EMG with enhanced synchronous input, suggests higher levels of MU synchrony within and between muscles in PD.The simulation results additionally suggest that this could be caused by an increase in synchronous inputs to the motoneuron pool in the beta frequency range.

Disclosures: **M.W. Flood:** A. Employment/Salary (full or part-time):; Science Foundation Ireland - Insight Centre for Data Analytics, University College Dublin. **B.R. Jensen:** A. Employment/Salary (full or part-time):; Odense University Hospital, University of Southern Denmark. **A. Malling:** A. Employment/Salary (full or part-time):; Odense University Hospital, University of Southern Denmark. **M.M. Lowery:** A. Employment/Salary (full or part-time):; University College Dublin. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Science Foundation Ireland - Insight Centre for Data Analytics, European Research Council - Consolidator Grant.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.09/OO19

Topic: E.10. Motor Neurons and Muscle

Support: European Union's Horizon 2020 research and innovation programme Marie Curie Individual Fellowship under grant agreement No #702491

Title: The role of common synaptic input to populations of motor neurons in the generation of force

Authors: *F. NEGRO[1], K. G. KEENAN[2], C. ORIZIO[1]
[1]Univ. degli Studi di Brescia, Dept. of Clin. and Exptl. Sci., Brescia, Italy; [2]Human Movement Sci., Univ. of Wisconsin-Milwaukee, Milwaukee, WI

Abstract: Spinal motor neuron activity is modulated by a combination of shared and independent synaptic input from spinal and supraspinal regions of the central nervous system. For this reason, correlation analysis between populations of motor neuron spike trains recorded *in vivo* shows a significant amount of synchronization between action potential discharge timings, both in low and high frequency bands. It has been hypothesized that low frequency components (< 5 Hz) are the effective neural drive to muscle and the main determinants of force control. On the other hand, high frequency components (6-32 Hz) may have a role in the generation of fast movements, tremogenic oscillations, and non-linear force control. Even at very low forces, the averaging process performed by the activation of hundreds of motor neurons act as a linear filter that attenuates uncorrelated synaptic activity and transmits the shared components almost unchanged to the force output. For these reasons, it is important to understand which characteristics of the shared synaptic input may influence the force signal and the functional significance of correlated motor neuron activity in human motor control. In this study, we begin to address these questions using a computational model comprising 445 motor units, similar to the tibialis anterior muscle, that received a broad frequency Gaussian synaptic input (0-50 Hz) with different levels of synaptic activations and shared input (0-100%). The first set of simulations showed that the amount of common synaptic input has a moderate influence on the rate-of-force-development at relatively low force levels and almost no impact at higher forces. On the other hand, changes in the amount of common synaptic input to the motor neuron pool had a significant effect on the linearity of the relation between variability and mean simulated force. Specifically, the inclusion of the shared synaptic input was necessary to increase the slope of the relation between standard deviation (SD) and mean force level after full motor unit recruitment (0-75% MVC) to better approximate experimental findings. Similar behavior was observed in the simulation of a smaller muscle (120 motor units, as in the first dorsal

interosseous of the hand) with a narrower recruitment range (0-50% MVC). In conclusion, the likely functional role of common synaptic input to populations of motor neurons innervating a single muscle is to shape the variability of the force output, but not to increase the rate of force development in fast contractions.

Disclosures: F. Negro: None. **K.G. Keenan:** None. **C. Orizio:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.10/OO20

Topic: E.10. Motor Neurons and Muscle

Support: DARPA Grant N66001-16-2-4052

NSF Grant 1527202

DHHS/NIDILRR Grant 90IF0064

DOD Grant OR140147

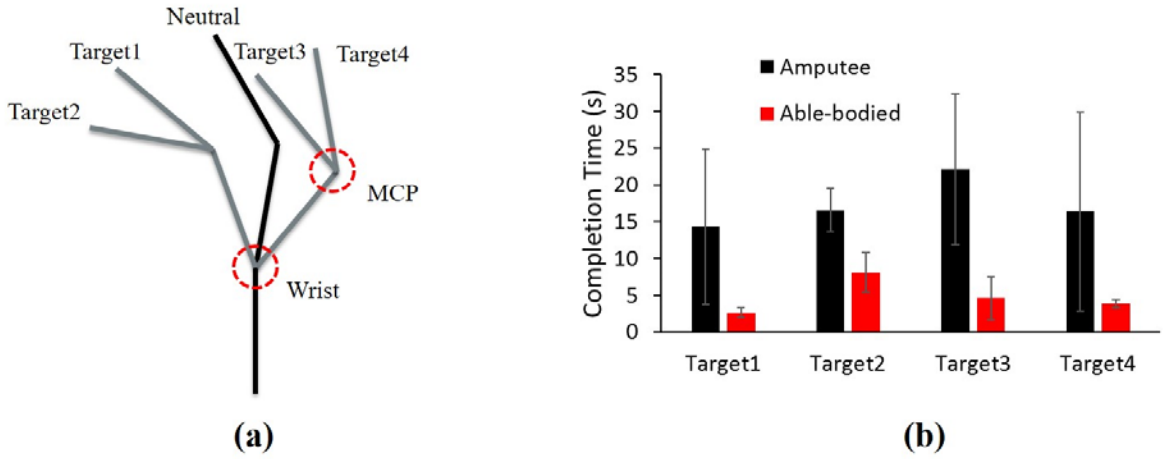
DOD Grant 13014002

Title: Can one EMG-based neural-machine interface fit all

Authors: *L. PAN, D. CROUCH, H. HUANG
NCSU/UNC Joint Dept. of Biomed. Engin., NC State Univ. and UNC-Chapel Hill, Raleigh, NC

Abstract: Introduction: Electromyography (EMG)-based movement intent decoding for neural-machine interfaces (NMI) has been used for prosthesis control for several decades. No matter which algorithm is used for EMG decoding, customizing the NMIs for individual users is always needed. Recently, we developed an EMG decoder based on a musculoskeletal model for predicting simultaneous flexion/extension motions at the metacarpophalangeal (MCP) and wrist joints. Since the model directly incorporates biological movement processes, and humans' own neural control is adaptive, customizing the model for each user might not be necessary. Therefore, the objective of this study was to investigate how well previously uninvolved subjects could control a generic EMG-based NMI in real-time to perform a task.
Materials and Methods: Based on previous data recorded from 6 able-bodied subjects, we developed a generic musculoskeletal model by averaging the model parameter values across all subjects and all trials. The model was used to control the position of a virtual hand on a computer screen. One other male able-bodied subject and one female transradial amputee were instructed to move a planar virtual hand from a Neutral posture to four Target postures (Fig. 1.a). The four

targets were presented in a random order in each of 4 trials. EMG were recorded from the able-bodied subject from surface EMG electrodes placed over four muscles: ECRL, ED, FCR, and FD. For the amputee subject, four fine-wire EMG electrodes were inserted in the same four muscles under ultrasound guidance.



Results: Impressively both the able-bodied and the amputee subjects completed all trials for all target postures, although the completion time of the able-bodied subject was lower than that of the amputee subject for all Target postures (Fig. 1.b).
Discussion and Conclusions: The results suggest that a generic EMG-based NMI could potentially enable effective real-time NMI control for both able-bodied subjects and arm amputees. More training might be needed to further improve amputee performance in using EMG-based NMI for prosthesis control.

Disclosures: L. Pan: None. **D. Crouch:** None. **H. Huang:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.11/OO21

Topic: E.10. Motor Neurons and Muscle

Support: ERC-2014-CoG-646923

Title: Influence of muscle architecture on the sEMG signal of the first dorsal interosseous muscle: A subject-specific model based on diffusion tensor imaging

Authors: *D. PEREIRA BOTELHO[1], N. COLGAN[2], A. FAGAN[3], K. CURRAN[1], M. LOWERY[1]

[1]Univ. Col. Dublin, Dublin, Ireland; [2]Natl. Univ. of Ireland, Galway, Ireland; [3]Trinity Col. Dublin, Dublin, Ireland

Abstract: The aim of this work is to examine the influence of the muscle architecture on the surface electromyography (sEMG) signal recorded during the activation of the first dorsal interosseous (FDI), a multifunctional muscle of the hand responsible for both index finger flexion and abduction. Diffusion tensor imaging (DTI) was used to assess fiber track information, which was incorporated into a subject-specific finite element model of the electric potential detected at the skin surface. General anatomical features of the hand were assessed through conventional magnetic resonance imaging (MRI). Subjects were positioned lying prone in a Philips 3T Achieva MRI scanner with their dominant hand prone (palm down) and supported on a purpose-built rig in a 16-element transmit/receive radiofrequency knee coil. The MRI compatible rig was conceived to minimize involuntary hand motion and provide loads for index flexion and abduction. DTI data were acquired using the same experimental arrangement with the hand at rest and while the subjects performed index finger abduction and flexion. A deterministic fiber tracking approach was used, after motion and eddy current correction, to estimate FDI muscle fiber direction. The acquired MRI data for the hand at rest were segmented into distinct tissues (bone, muscle, fat, skin) and a three-dimensional model of the hand was constructed. The finite element model was then created with different dielectric properties associated with each tissue. The estimated muscle fiber direction and curvature were used to determine material anisotropy and the trajectory of action potential propagation. Single fiber action potentials at an array of electrodes located on the skin surface above the FDI were then estimated as the convolution of a weighting function representing the response at the electrode due to propagation of a unit point current source and the intracellular action potential current, incorporating fiber start- and end-effects at the neuromuscular junction and tendon. Motor unit action potentials comprised of single fiber action potentials from all fibers within the motor unit were then simulated during index finger flexion and abduction. The proposed approach constitutes a basis for simulation of subject-specific muscle models which can be used to investigate muscle activation under a range of conditions. Insights obtained through a better understanding of the contribution of structural and architectural properties of the muscle and surrounding tissues enable the extraction of precise/specific information regarding the state of the muscle and nervous control mechanisms using advanced sEMG recording and analysis techniques.

Disclosures: D. Pereira Botelho: None. **N. Colgan:** None. **A. Fagan:** None. **K. Curran:** None. **M. Lowery:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.12/OO22

Topic: E.10. Motor Neurons and Muscle

Title: High-density electromyogram decomposition using different blind source separation algorithms

Authors: *C. DAI, X. HU
Univ. of North Carolina At Chapel Hill, Chapel Hill, NC

Abstract: Electromyogram (EMG) decomposition is a technique that can separate the composite EMG signals into constituent motor unit spike trains. Blind source separation algorithms are efficient approaches that can decompose high-density EMG signals recorded from a densely-spaced electrode grid. Our study compared three independent component analysis (ICA) based blind source separation algorithms (i.e., InfomaxICA, FastICA and RobustICA) in combination with the convolution kernel compensation method. We also examined the overall decomposition performance of these different approaches in both offline and online configurations. These algorithms were evaluated using 64-channel simulated EMG signals with different numbers (10, 20 and 50) of motor units and signal-to-noise ratios (5, 10 and 20 dB). The performance of each algorithm was evaluated based on the yield of the detected motor units and the spike timing accuracy of individual motor units. Our offline decomposition results showed that all algorithms had comparable performance with mean accuracy > 98% for the simulated EMG signals with 10 or 20 motor units. However, the RobustICA outperformed (in both the yield and timing accuracy) the FastICA and the InfomaxICA for more complex signals with 50 motor units. In addition, the computation time of the RobustICA was approximately 25% shorter than the FastICA, and both algorithms were substantially faster than the InfomaxICA. The real-time decomposition of RobustICA and FastICA was also performed on the simulated EMG with 50 motor units. The accuracy was approximately 90% and approximately 7 motor units were detected by using RobustICA or FastICA. Our findings indicate that the RobustICA-based decomposition technique can provide accurate decomposition results in both offline and real-time settings. Our approach shows great promise as a research tool that can help us understand the control of motoneuron pool. The online decomposition outcome can also be used as a real-time control input of rehabilitation/assistive devices, which can help improve system performance.

Disclosures: **C. Dai:** None. **X. Hu:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.13/OO23

Topic: E.10. Motor Neurons and Muscle

Title: Development of a simple device for assessment of spasticity

Authors: *K. TAKEDA[1], H. MAEDA[2], S. SONODA[2]
[1]Sch. of Hlth. Sci., Fujita Hlth. Univ., Toyoake, Aichi, Japan; [2]Dept. of Rehabil. Med. II, Sch. of Med., Fujita Hlth. Univ., Tsu, Mie, Japan

Abstract: Stroke, spinal cord injuries, multiple sclerosis, cerebral palsy, and traumatic brain injury can cause spasticity, which is an involuntary and abnormal muscle tone. Spasticity causes difficulty in motion, abnormal posture, and contractures leading to limited joint mobility. Clinical evaluation of spasticity has been usually performed by modified Ashworth scale (MAS) which is an ordinal scale that assesses the resistance during passive joint movement by an examiner's subjectivity. In the present study, for an objective assessment, we developed a simple device for an ankle joint spasticity, which measures resistance torque (plantar flexion torque) during manual passive dorsiflexion. The device consists of an ankle-foot orthosis (AFO) that can move the ankle joint from 25 degrees of plantar flexion to 15 degrees of dorsiflexion. The resistance torque is measured by strain gauges which were attached to the vicinity of the toe of the AFO. Simultaneously, the joint angle can be measured by a potentiometer which is installed on the ankle joint axis. This proposed system is also be able to record surface electromyography (sEMG) of the gastrocnemius (GC) and tibialis anterior (TA) muscles simultaneously at 1 kHz sampling. The time course of the torque, angle, angular velocity, and rectified sEMG signals is drawn with a slight time delay on a PC display. Because a generally accepted definition of spasticity is ‘a velocity dependent increase in tonic stretch reflex with exaggerated tendon jerks, resulting from the hyperexcitability of the stretch reflex’, it is important to dorsiflex the ankle joint passively in the same angular velocity. Therefore, the proposed device also provides two guides so that the examiner can perform dorsiflex at a constant angular velocity. For one of the guides, the time course of the angle that the examiner should trace is indicated visually on the display in advance. Another one is sound guide which is an up-chirp signal with starting frequency 440 Hz and ending frequency 880 Hz of a duration of the dorsiflexion time. For the dorsiflexion of the ankle joint at a low angular velocity, e.g. under 2 deg/s to assess an elasticity of the joint, high reproducibility could be seen by relying on the visual guide. On the other hand, high reproducibility at a high angular velocity over 200 deg/s could also be performed by using the auditory guide. The proposed system may be useful for assessing spasticity objectively.

Disclosures: K. Takeda: None. **H. Maeda:** None. **S. Sonoda:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.14/OO24

Topic: E.10. Motor Neurons and Muscle

Title: Sources of muscle weakness in stroke survivors based on a motor unit simulation

Authors: *X. HU[1], H. SHIN[1], W. Z. RYMER[2], N. L. SURESH[3]
[1]BME, UNC Chapel Hill, Chapel Hill, NC; [2]Rehabil. Inst. of Chicago, Chicago, IL; [3]Sensory Motor Perf Prgm, Rehabil. of Chicago, Chicago, IL

Abstract: Muscular weakness is a major motor impairment in stroke survivors. Concurrent impairment in the control of motor unit pool can contribute to the observed weakness. However, the relative contribution of different aspects of impaired motor unit control is not clear. To address this issue, we used a physiologically based motor unit pool model to simulate different altered motor unit properties observed experimentally in stroke survivors. Our results showed that a reduction in the discharge rate and the motor unit size (i.e., twitch amplitude) led to substantial muscle weakness. Additionally, a reduced recruitment range (i.e., a clustered recruitment at a low excitation drive) also contributed to muscle weakness substantially. In contrast, a disturbance in the discharge rate organization across units had moderate influence on the force output, and a disturbance in the orderly recruitment (i.e., size principle) had a minimal impact on muscle weakness. Our findings indicate that several factors can contribute to muscle weakness, including an inefficient activation of the motor units at low discharge rate, a reduced motor unit size possibly due to atrophy, and a compressed recruitment threshold of motor units triggering early muscle fatigue. Overall, using a previously developed motor unit pool model, we were able to distinguish the different altered motor unit properties in contribution to muscle weakness in stroke survivors.

Disclosures: X. Hu: None. **H. Shin:** None. **W.Z. Rymer:** None. **N.L. Suresh:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.15/OO25

Topic: E.10. Motor Neurons and Muscle

Title: Muscle synergies underlying sit-to-stand tasks in acute stroke patients

Authors: *H. HANAWA[1], K. KUBOTA[1], T. KOKUBUN[2], K. HIRATA[1], T. MIYAZAWA[3], M. SONOO[1], T. FUJINO[1], N. KANEMURA[2]
[1]Grad. Course of Hlth. and Social Services, Grad. Sch. of Saitama Prefectural Univ., Koshigaya, Japan; [2]Dept. of Hlth. and Social Services, Saitama Prefectural Univ., Koshigaya, Japan; [3]Ageo Futatsumiya Clin., Ageo, Japan

Abstract: *[INTRODUCTION]* Neurophysiological evidence suggests that the nervous system controls motion by using a low-dimensional synergy organization for muscle activation.

Recently, abnormal muscle synergies in stroke patients have also been clarified. However, it is not clear if such abnormal muscle synergies occur during sit-to-stand tasks.
[METHODS] Three stroke patients with left hemiparesis (time since stroke = 6.3 ± 2.3 days), and two healthy adults participated in this study. A surface electromyogram (EMG) device was used to collect muscle activity data from seven muscles (tibialis anterior, soleus, gastrocnemius, vastus lateralis, rectus femoris, long head of biceps femoris [BFL], and gluteus maximus [GM]) in the left leg. Subjects performed a six-direction isometric contraction task in the supine position to verify fundamental muscle synergies. Then, they performed a sit-to-stand task. A non-negative matrix factorization algorithm was applied to the EMG data for each task to extract muscle synergies. Subsequently, hierarchical cluster analysis was used to classify these synergies.
[RESULTS] Four subjects accomplished all isometric contraction tasks with four muscle synergies. Each muscle synergy was included in the same cluster (correlation coefficient = 0.84 ± 0.06). One patient with severe hemiparesis could perform only one-sixth of the contraction directions. The number of muscle synergies of this patient decreased to two. All subjects completed the sit-to-stand task. Healthy adults had four muscle synergies. In two of the stroke patients, the number of muscle synergies decreased to three as GM activity disappeared. To compensate, these patients prolonged the BFL activity; therefore, this muscle synergy formed a different cluster from healthy adults. The patient with severe hemiparesis had four muscle synergies. Because most muscles are co-active in the extension phase, this muscle synergy formed a different cluster from healthy adults. Other muscle synergies were included in the same cluster in all subjects (correlation coefficient = 0.93 ± 0.01).
[DISCUSSION] The presence of fewer muscle synergies in stroke patients corresponds to an overall reduction in motion control complexity (Bowden MG, 2010). This finding was applicable to the sit-to-stand task. However, muscle synergies in stroke patients were different in the two tasks. Patients likely changed fundamental muscle synergies to achieve the sit-to-stand task, even in the acute phase. This study contributes to the findings regarding how a nervous system injury may alter the organization of muscle synergies.

Disclosures: H. Hanawa: None. **K. Kubota:** None. **T. Kokubun:** None. **K. Hirata:** None. **T. Miyazawa:** None. **M. Sonoo:** None. **T. Fujino:** None. **N. Kanemura:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.16/OO26

Topic: E.10. Motor Neurons and Muscle

Title: Relationship between muscle synergy in knee osteoarthritis patients and the functional evaluation of knee joints

Authors: *K. KUBOTA[1], H. HANAWA[1], T. KOKUBUN[2], M. SONOO[1], K. HIRATA[1], T. FUJINO[1], G. OONO[1], N. KANEMURA[2]
[1]Grad. Course of Hlth. and Social Services, Grad. Sch. of Saitama Prefectural Univ., Koshigaya/Saitama, Japan; [2]Saitama Prefectural Univ., Koshigaya/Saitama, Japan

Abstract: *[Introduction]*Muscle synergy is believed to reflect alterations in motion control caused by a dysfunction in the redundant musculoskeletal system. While muscle synergy has been applied to central nervous system diseases, there are still few applications for orthopedic diseases. Knee osteoarthritis (KOA) is a degenerative disease that features osteophyte formation, cartilage destruction, and joint space narrowing. KOA is thought to have sensory and motor impairments that affect force generation and motor unit recruitment of the knee extensors and flexors. While several previous studies have shown changes in individual muscle activity, there have been no studies to show changes in muscle synergy. The purpose of this study was to clarify the relationship between changes in muscle synergy and knee function evaluation in KOA.
*[Methods]*Six healthy adults and three KOA patients participated in this study. Subjects performed gait on the treadmill at different speeds (1km/h or 2 km/h). EMG activity was recorded from 14 left leg and lower-back muscles. The gait cycle was defined using ground reaction force data obtained from each belt of the treadmill. Data were time-interpolated over individual gait cycles to fit a normalized 200-point time base. EMG signals were factored to apply non-negative matrix factorization to determine the muscle synergy. To determine the minimum number of synergy, we calculated the variability accounted for. Furthermore, to compare knee function evaluation, the following outcomes were evaluated: knee pain of WOMAC and range of motion (ROM) of both knees.
*[Results]*Healthy subjects had extracted muscle synergy values of three or four, while KOA patients had extracted muscle synergy value of two. Synergy during the stance phase was confirmed using simultaneous activity of the lower leg and multiple thigh muscles in KOA patients. The extension ROM was -5° for KOA1, -10° for KOA2, and -10° for KOA3. The WOMAC scores of the three KOAs were, on average, 37/96.
*[Discussion]*The main finding of this study was that muscle synergy was reduced in KOA patients. A reduction in muscle synergy indicates the co-activation of muscles. As many previous studies have indicated, co-activation of muscles results in increased joint stiffness. In this way, muscle synergy can be manipulated to address the pain and instability of KOA patient. Our results support these findings. The abnormal structuring of muscle synergy may reflect both abnormal musculo-skeletal dynamics and other biomechanical properties of the limb. This study contributes to the finding that musculo-skeletal system injury may alter the organization of muscle synergies.

Disclosures: K. Kubota: None. **H. Hanawa:** None. **T. Kokubun:** None. **M. Sonoo:** None. **K. Hirata:** None. **T. Fujino:** None. **G. Oono:** None. **N. Kanemura:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.17/OO27

Topic: E.10. Motor Neurons and Muscle

Title: Anterior interosseous nerve syndrome variant confirmed by MRI and electromyography

Authors: *I. JUNG[1], S. JEE[1], I. KIM[2]
[1]Dept. of Rehabil. Med., Chungnam Natl. Univ. Hosp., Daejeon, Korea, Republic of; [2]Dept. of Rehabil. Med., Seoul Natl. Univ. Col. of Med., Seoul, Korea, Republic of

Abstract: Introduction: Anterior interosseous nerve (AIN) syndrome is a rare disorder with complex clinical presentations. Anatomical variation of the proximal median nerve has been reported and should be confirmed before identifying the lesion of AIN.
Methods: We report two similar cases of an AIN variant. Patients presented with pain sensations and subsequent hand weakness. To confirm muscle denervation and identify the causes of proximal median nerve injury at the forearm, electromyography and magnetic resonance imaging (MRI) were performed.
Results: The electrodiagnostic study suggested proximal median nerve injury and AIN. However, no abnormal nerve conduction in the median sensory nerve and abductor pollicis brevis was found. Denervation of muscles innervated by the proximal median nerve was observed in MRI.
Conclusion: We observed an AIN variant innervating muscles dominated by the proximal median nerve. We recommend that this be considered in future examinations to confirm lesions of whole median nerve fascicles through electromyography or MRI.

Disclosures: I. Jung: None. **S. Jee:** None. **I. Kim:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.18/OO28

Topic: E.10. Motor Neurons and Muscle

Title: Modulation of jaw reflex responses during peripherally and centrally evoked swallowing

Authors: *T. SUZUKI, M. YOSHIHARA, K. TSUJI, J. MAGARA, T. TSUJIMURA, M. INOUE
Div. of Dysphagia Rehabil., Niigata Univ. Grad. Sch. of Med. and, Niigata-Shi, Japan

Abstract: [Purpose] We previously demonstrated that the jaw-opening reflex (JOR) responses were inhibited during swallowing evoked by superior laryngeal nerve (SLN) stimulation. The aim of this study was to investigate whether the JOR responses were modulated during not only peripherally but also centrally evoked swallowing. We also examined the effect of peripherally and centrally evoked swallows on the jaw-closing reflex (JCR) responses. [Materials and Methods] Experiments were carried out on rabbits anesthetized with urethane. Electromyographic activity was recorded from the digastric, masseter and thyrohyoid muscles for monitoring the JOR, JCR and swallowing, respectively. To evoke the JOR and JCR, the inferior alveolar nerve and the trigeminal mesencephalic nucleus were electrically stimulated. The intensity of the stimulus pulse was 2.0 times (T) the threshold for eliciting the JOR and JCR. Either the SLN or cortical swallowing area (Cx) was stimulated at 30 Hz to evoke swallows. Stimulus intensity of SLN and Cx was set at 0.8-4 T and 0.8-1.4 T the threshold for evoking the swallowing at least once for 10 sec, respectively. In a recording session, either the JOR or JCR was evoked continuously at 1 Hz for 30 sec. In the middle 10 sec, SLN or Cx was simultaneously stimulated. The mean amplitudes and latencies of the JORs and JCRs were compared among the conditions; before, during and after SLN or Cx stimulation. [Results & Discussion] The amplitudes of the JORs were significantly decreased during and after SLN/Cx stimulation compared with before stimulation, whereas the latencies were not changed among the conditions. Inhibitory effect increased as the stimulus intensity of the SLN and Cx increased. On the other hand, there was no significant difference in the amplitudes and the latencies of the JCRs among the conditions. These results suggest that the JOR was inhibited, whereas the JCR was not modulated during peripherally and centrally-evoked swallowing, probably to prevent unnecessary jaw opening evoked by the oral sensory inputs during swallowing.

Disclosures: **T. Suzuki:** None. **M. Yoshihara:** None. **K. Tsuji:** None. **J. Magara:** None. **T. Tsujimura:** None. **M. Inoue:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.19/OO29

Topic: E.10. Motor Neurons and Muscle

Title: The influence of the difference in the volume of keying on trunk movement during piano performance

Authors: *A. KOBAYASHI[1], T. KOKUBUN[2], D. NOHARA[3], H. SHONO[3], M. MATSUNO[3], H. HANAWA[4], N. KANEMURA[2]
[1]Grad. Sch. of Hlth. and Social Services, Saitama Prefectural Univ., Koshigaya/Saitama, Japan; [3]Fac. of Hlth. and Social Services, Dept. of Physical Therapy, [2]Saitama Prefectural Univ., Koshigaya City, Saitama, Japan; [4]Grad. Course of Hlth. and Social Services, Grad. Sch. of Saitama Prefectural Univ., Koshigaya, Japan

Abstract: INTRODUCTION: Pianists acquire sophisticated performance skills through years of practice. As with other sportsmen, they often have somatic complaints. Recently, incorrect usage of the body was referred to as one of the causes of this disorder. Compared to the old piano, the modern piano requires approximately five times the keying force; therefore, the pianists are required to play rationally with the whole body. The aim of this study is to clarify the role of the trunk during the playing of the piano, and it is shown that the role of the trunk can be effectively used for the prevention and treatment of a pianist's disorder. METHOD: Five pianists who have received piano instruction for 20 ± 12.7 years and five non-pianists participated in the study. By using the 3D motion-analysis device VICON, the motion of the trunk, upper limbs, and fingers during a key stroke was recorded. The conditions of sound intensity were 64 and 96 MIDI velocity. For the musical performance, the accuracy of the key depression tempo and the keying volume and its coefficient of variation were calculated. In addition, the keying contribution degree and keying contribution rate were calculated from the measured trunk inclination angle and the joint angles of the shoulder, elbow, wrist and finger. The Friedman test was carried out and the Wilcoxon signed-rank sum test was conducted for multiple comparisons. All experimental protocols were reviewed and approved by the Research Ethics Review Committee of Saitama Prefectural University. RESULTS: Both sets of pianists and non-pianists could key at the prescribed tempo and volume; the coefficient of variation of the keying volume was smaller when the volume was higher. When the volume was small, the pianists showed a higher positive contribution. Further, as the volume increased, the positive contribution of both the pianists and non-pianists decreased. The pianists showed higher keying efficiency at any given volume and displayed an increase in the keying efficiency as the volume increased, but the non-pianists portrayed a decrease in the keying efficiency as the volume increased.DISCUSSION: The pianist was using the trunk to hit the key at a small volume. It was more difficult to hit a key with a small volume. Pianists took a strategy to shorten the distance between the trunk and the piano against a challenging task. It is suggested that pianists play not only with arms and hands, but also trunks. The results obtained by measuring the movement of the entire body during a piano performance suggest the necessity of such an evaluation, with special emphasis on the trunk.

Disclosures: A. Kobayashi: None. **T. Kokubun:** None. **D. Nohara:** None. **H. Shono:** None. **M. Matsuno:** None. **H. Hanawa:** None. **N. Kanemura:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.20/OO30

Topic: E.10. Motor Neurons and Muscle

Title: Knee flexors and extensors are defined by the task and not anatomy

Authors: *P. SRIYA, T. RICHARDS, S. ASTILL, S. CHAKRABARTY
Sch. of Biomed. science, Fac. of Biol. Sci., Leeds, United Kingdom

Abstract: The knee is a modified hinge joint used for both flexion and extension thus allowing one to walk, stabilize posture and do sit to stand. Control of the knee is typically assigned to the flexors (semitendinosus (ST) and bicep femoris (BF)) and extensors (rectus femoris (RF), vastus lateralis (VL), vastus medialis (VM) and vastus intermedius(VI)). It is assumed that the activity at the knee is defined by that of these muscles, which maintain their respective role across all knee positions and range of motion. In this study, we examined this premise by recording surface EMGs from the flexors (ST and BF) and extensors (RF, VL and VM) simultaneously during an isometric knee extension task in 2 distinct positions across the range of motion. We hypothesized that the isometric conditions of the task will allow interactions between the muscles to be unaffected, thus remaining the same across conditions and range of motion (**0°, 20°, 60°** and **90°)**. In position 1, the participant was supine lying with both legs stretched forward, and in position 2, their left knee is bent towards the gluteal on the bed.We examined the activation patterns (onset, offset and duration) along with RMS amplitude changes in these muscles. Using hierarchical clustering and Pearson's correlations across 17 healthy participants (18-30), we found that in at **0°** and **20°** in position 1 RF and VL acted as putative agonists. At **60°** in position 1 and at **90°** in position 2 the activity patterns were similar with RF, VL, ST and BF in the same agonist cluster, but not VM. As if VM in this scenario is more a flexor than its traditional role of an extensor. It is likely, as has been previously suggested that the muscles do undergo stretching thus providing proprioceptive feedback. We suggest that the muscle interactions are controlled dynamically and not anatomically regulated, possibly by the proprioceptive feedback from muscles and joints. This has significant implications for clinicians, as these tests are routinely used to assess functional state of the knee, but the variations observed suggest the 'normal' needs to be redefined.

Disclosures: P. Sriya: None. **T. Richards:** None. **S. Astill:** None. **S. Chakrabarty:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.21/OO31

Topic: E.10. Motor Neurons and Muscle

Support: NIH NINDS 5P01NS055976

Title: Concurrent recordings of motor unit and spinal interneuron discharge patterns during reflex activation

Authors: *C. K. THOMPSON[1], F. MARCHIONNE[2], A. J. KRUPKA[2], F. NEGRO[3], D. FARINA[4], M. A. LEMAY[2]
[1]Spinal Neuromotor Lab., [2]Dept. of Bioengineering, Temple Univ., Philadelphia, PA; [3]Dept. of Clin. and Exptl. Sci., Univ. degli Studi di Brescia, Brescia, Italy; [4]Dept. of Bioengineering, Univ. Col., London, United Kingdom

Abstract: Spinal alpha motoneurons receive most of their synaptic input through spinal interneurons, yet there are few investigations linking the population activity of spinal interneurons to the discharge of spinal motoneurons. Here we present an approach to record the discharge times of several tens of spinal interneuron in combination with motor units in order to better quantify how interneurons influence the firing of spinal motoneurons. The experiment was performed using the spinal decerebrate cat. A laminectomy was performed from L3 through L6, dura and pia over the recording sites were removed. The right hind limb was secured to a rigid frame and the posterior leg muscles were exposed. Two separate 64-channel intraspinal linear microelectrode arrays were inserted at L3 and L6 just medial to the ipsilateral dorsal root entry zone to record throughout the intermediate zone of the spinal gray. Additionally, a single 64-channel electrode array was placed onto the muscle belly of the right medial gastrocnemius muscle. Reflexive motor activity was evoked through an eccentrically weighted rotary motor rotating at ~100Hz that was held manually against the distal tendon of the right medial gastrocnemius for 30 seconds. Neural and EMG data were decomposed offline into their corresponding discharge times. Discharge times from 50-70 interneurons and 10-15 motor units were decomposed from each trial. Motor units from the medial gastrocnemius were quiescent at rest, but showed a robust increased discharge rate in response to vibratory stimulus, each motor unit spike train demonstrates an ISI histogram that was highly punctuated at subharmonics of the vibration frequency. In contrast, a large majority of interneurons were active at rest. During vibration, interneurons demonstrated a significant increases in average discharge rate (from 11 to 19 imp/s), though less than 10% of the recorded interneurons demonstrated punctuated ISI histograms. These data are consistent with the direct monosynaptic projections from the muscle spindle onto the homonymous motor pool; highly punctuated ISI histograms are exclusively

observed at the level of the motoneurons, but observed relatively infrequently across spinal interneurons. This approach may be helpful in identifying the synaptic input of specific spinal interneurons. Moreover, the ability to collect these spike train data will allow us to better quantify the synaptic input from spinal interneurons to spinal alpha motoneurons under a variety of reflexive, pharmacological, and surgical conditions, further elucidating the role of spinal interneurons on motor output.

Disclosures: C.K. Thompson: None. **F. Marchionne:** None. **A.J. Krupka:** None. **F. Negro:** None. **D. Farina:** None. **M.A. Lemay:** None.

Poster

412. Motor Unit Recordings

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 412.22/OO32

Topic: E.10. Motor Neurons and Muscle

Title: Sex differences in neuromuscular control of quadriceps muscles during various levels of knee extension force

Authors: *Y.-L. PENG, D. GUPTA, J. JENSEN, L. GRIFFIN
Dept. of Kinesiology and Hlth. Educ., The Univ. of Texas At Austin, Austin, TX

Abstract: Patellofemoral pain syndrome is twice as prevalent in women than in men. The main cause of increased rates in women is believed to be due to imbalance of control of muscles tracking the patella. Clinicians commonly recommend vastus medialis oblique (VMO) training to counteract the excessive force of the vastus lateralis (VL) on the patella. However, it is unclear whether the activation patterns of the vastus muscles differ between sexes and force levels. The purpose of the study is to investigate the onset time and EMG amplitude of the quadriceps muscles in healthy individuals during isometric knee extension at three force levels. Thirteen men and 13 women performed isometric knee extension at 25%, 50%, and 75% of maximal voluntary contraction (MVC). All participants performed a ramp contraction by tracing a line on the screen with a rate of rise of 7.5% MVC/sec up to the target force. Levels were maintained for 10 sec at 25% and 50% MVC and for 5 sec at 75% MVC. Surface electromyography (EMG) was recorded from the VL, VMO, vastus medialis (VM), and rectus femoris (RF) muscles. EMG onset time and average holding EMG amplitude (normalized to maximum) were determined. A two-way repeated measure ANOVA with Tukey post hoc tests were used to compare onset time between Sex (male/female) and Muscle (VM/VMO/VL/RF). EMG amplitude was evaluated across Force (25/50/75%MVC), Sex, and Muscle by a three-way repeated measure ANOVA with Tukey post hoc tests. There was a significant Muscle effect on the onset time (p=0.00). The VL (0.21±0.04 sec) was activated significantly earlier than the VM (0.47±0.04 sec) and RF (0.56±0.07 sec), but not the VMO (0.37±0.06 sec). There was a significant 3-way interaction of

Muscle*Sex*Force for the EMG amplitude (p=0.04). The activation amplitude of the VL (21±5% EMGmax) was significantly lower than the VM (46.6±5% EMGmax) and RF (55.5±8% EMGmax), but was not different than the VMO (36.9±9% EMGmax). Women showed 36.7±8% EMGmax higher percentage of muscle activation for all the vastus muscle compared to men to complete the task in all force levels (p=0.00). A marginal significant difference in the EMG amplitude was found between the 25% and 75% force level (p=0.06). Different force levels had less effect on the activation pattern of the quadriceps muscles. However, the VL and VMO were recruited earliest muscles, possibly to counterbalance the lateral and medial force on patella during the knee extension task. With the same relatively force exertion, women exhibited much higher muscle relative muscle activation of all vastus muscles compared to men.

Disclosures: Y. Peng: None. **D. Gupta:** None. **J. Jensen:** None. **L. Griffin:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.01/OO33

Topic: F.04. Stress and the Brain

Title: Stress-induced pro- and anti-inflammatory cytokine expression in casual drug and alcohol users

Authors: *M. ARCHEY[1], K. N. SHEFFIELD[1], C. NEUTZLER[1], H. R. RHODES[1], J. A. BOYETTE-DAVIS[2]
[2]Psychology, [1]St. Edward's Univ., Austin, TX

Abstract: Research shows that various recreationally used substances have distinctive effects on neuromodulation, and those effects can produce observable health outcomes. For example, while alcohol is associated with increased inflammation in the body, marijuana has been shown to possess anti-inflammatory properties. The present study aimed to investigate how recreational use of these substances and their corresponding modulation of cytokines, in conjunction with daily, non-illness related stress, can influence overall health and well-being. Seventy-nine participants completed sessions in which they provided detailed information about their health, frequency of symptoms associated with inflammation, self-reported stress, personality, and drug and alcohol use. Based on self-reports, participants were categorized as abstainers (no reported drug/alcohol use), daily users of marijuana, marijuana plus alcohol users, moderate alcohol users, or heavy/binge alcohol users. During the session, participants provided saliva samples before and after exposure to mild, acute stress, induced by the completion of both mental arithmetic and Stroop tests. Inflammation scores were significantly correlated with perceived stress ($r = .48$, $p = .003$). Cytokine levels - interleukin-6 (IL-6) and interleukin-10 (IL-10) - were analyzed via saliva by enzyme immunoassays. No significance was found for IL-10 levels. However, results

revealed significant increases in IL-6 according to differential substance use patterns. Specifically, those who were daily consumers of marijuana or moderate users of alcohol experienced increases in this pro-inflammatory cytokine. This study highlights the importance of investigating immune function in recreational drug users and the overall effect of controlled substances on health and stress.

Disclosures: M. Archey: None. **K.N. Sheffield:** None. **C. Neutzler:** None. **H.R. Rhodes:** None. **J.A. Boyette-Davis:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.02/OO34

Topic: F.04. Stress and the Brain

Support: VA Merit Award BX002558-01

Title: COMTval158met polymorphism effects on inflammation and enduring stress response: A novel mechanism of PTSD risk

Authors: *J. DESLAURIERS[1,2], X. ZHOU[1,2], V. B. RISBROUGH[1,2]
[1]Psychiatry, Univ. of California San Diego, LA Jolla, CA; [2]Ctr. of Excellence for Stress and Mental Health, VA, La Jolla, CA

Abstract: Background and hypothesis: The catechol-*O*-methyltransferase (COMT) enzyme is implicated in the catabolism of catecholamines and plays a key role in cortical signaling. The val158met single nucleotide polymorphism (SNP) in the COMT gene has been associated with a greater risk for posttraumatic stress disorder (PTSD). Recent evidence has implicated immune function as a potential mechanism of PTSD risk and symptom maintenance. Because catecholamines play a key role in immune signaling we hypothesized that the COMTval158met SNP association with PTSD risk may be via alteration of the inflammatory state. We have previously shown, in a mouse line "humanized" for the COMTval158met SNP, that Val/Val carriers, compared to Met/Met carriers, exhibited altered inflammatory responses and greater enduring anxiety-like responses up to 14 days after predator stress exposure. To determine if greater inflammation plays a causal role in the enduring effects of stress in COMT mice, here we examined the long-term effects of inflammation on anxiety-like behaviors.
Methods and results: We investigated the effect of lipopolysaccharide (LPS), a toll-like receptor 4 (TLR4) agonist, in the "humanized" COMTval158met mouse line. LPS (1 mg/kg, IP) or saline was administered in male Met/Met and Val/Val carriers. Avoidance behaviors were assessed in the open field (exploratory behavior) and light-dark box (anxiety behavior) one week after injection. A composite avoidance score (Z score) was calculated for each animal across both

tests. We found that LPS increased avoidance behaviors in both genotypes, with the highest response in Val/Val mice. Val/Val mice also exhibited greater baseline inflammation compared to Met/Met mice.
Conclusions: These results (1) show that the COMTval158met SNP modulates the response to LPS; and (2) confirm that the COMTval158met-modulated inflammation plays a role in stress-induced behaviors. These findings suggest that targeting the immune signaling pathways might be a novel therapeutic alternative for PTSD patients, and that the COMTval158met SNP might modulate the response to anti-inflammatory treatments. The long-term effects of LPS on inflammatory markers in the "humanized" COMTval158met mouse line will also be presented.

Disclosures: J. Deslauriers: None. **X. Zhou:** None. **V.B. Risbrough:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; VA Merit Award BX002558-01.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.03/OO35

Topic: F.04. Stress and the Brain

Support: NIMH Intramural Research Program, ZIA MH001090

Title: Adaptive immunity in depressive mood states

Authors: *S. L. KIGAR[1], M. UDAWATTA[1], M. L. LEHMANN[1], S. J. LISTWAK[1], A. G. ELKAHLOUN[2], D. MARIC[3], M. HERKENHAM[1]
[1]Section on Functional Neuroanatomy, Natl. Inst. of Mental Hlth., Bethesda, MD; [2]Microarray Core, Natl. Human Genome Res. Inst., Bethesda, MD; [3]Flow and Imaging Cytometry Core, Natl. Inst. of Neurolog. Disorders and Stroke, Bethesda, MD

Abstract: Despite its striking prevalence in the general population, treatment options for most mood disorders including depression are quite limited; for example, available pharmacological agents fail to yield therapeutic value in up to 70% of patients diagnosed with major depressive disorder. Thus, novel treatment strategies and drug targets are urgently needed. Clinical and genome-wide association studies show a strong correlation between aberrant peripheral immunity and major depression in a subset of patients. Additionally, introducing cytokines (i.e., immune cell signaling products) into the body as a treatment for cancer in otherwise non-depressed patients was shown to induce depressive symptoms, suggesting that the immune system may play a role in the etiology of depression. These data and mounting evidence that normal immunity is critical for species-typical cognition and behavior have therefore engendered

increasing interest in elucidating how the immune system exerts influence on the brain. The relative impenetrability of the blood brain barrier (BBB) to immune cells and cytokines has led researchers to examine the immunogenic properties of the meninges, where lymphocytes of the adaptive branch of the immune system reside in abundance. Our laboratory recently showed that adoptive transfer of lymphocytes from chronically stressed donor mice conferred an anti-depressant effect in recipients (Brachman et al., 2015). This led us to hypothesize that (1) chronic stress alters the transcriptional profiles and signaling potential of T lymphocytes, and (2) that T cells within the meningeal compartment secrete a BBB-soluble factor that modifies a depressive-like behavioral phenotype. To investigate these hypotheses, we conducted microarray, flow cytometric, and immunohistological analyses to characterize T cells from a variety of immunological compartments, including the spleen, lymph nodes, and meninges. Moreover, using *in vitro* and *in vivo* assays, we explored the possibility that meningeal T cells release BBB-permeable, microRNA (miRNA)-containing extracellular vesicles, i.e., exosomes. Our preliminary data suggest that trafficking, activation status, and miRNA content of T cells is altered considerably by chronic social defeat stress, and these transcriptional changes impinge on the brain to affect mood.

Disclosures: S.L. Kigar: None. **M. Udawatta:** None. **M.L. Lehmann:** None. **S.J. Listwak:** None. **A.G. Elkahloun:** None. **D. Maric:** None. **M. Herkenham:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.04/OO36

Topic: F.04. Stress and the Brain

Support: CAPES

CNPq

FAPESP

Title: Inflammatory pathways in the medial prefrontal cortex modulate the anxiogenic effect induced by an acute stress

Authors: *S. F. LISBOA[1], C. VILA-VERDE[2], D. L. ULIANA[3], L. A. BRAGA[2], L. B. M. RESSTEL[4], F. S. GUIMARAES[2]
[1]Pharmacol., [3]Pharmacol. Departament, [4]Pharamacoloy, [2]Univ. of Sao Paulo - FMRP/USP, Ribeirao Preto, Brazil

Abstract: Background: Nitric oxide (NO) is involved in stress-related disorders and can be synthesized by three enzymes, including the inducible NO synthase (iNOS), which is involved in

the production of inflammatory mediators. Psychological stressors induce iNOS expression in cortical neurons and activate inflammatory mechanisms in the medial prefrontal cortex (MPFC), which is implicated in several psychiatric conditions. Nuclear factor kβ (NF-kβ) is the main regulator of iNOS expression and is also implicated in stress-related responses. However, it is unknown if MPFC inflammatory pathways, particularly iNOS and NF-kβ, exert key role in the development of behavioral consequences induced by stress. Therefore, the aim of this study was to determine if acute inhibition of iNOS or NF-kβ in the MPFC attenuated the anxiogenic-like effect induced by acute restraint stress (RS). Methods: Male Wistar rats (260-280 g) had guide cannulae implanted bilaterally in the MPFC for the administration of vehicle (0.2 µl), the selective iNOS inhibitor, N-[[3-(Aminomethyl)phenyl]methyl]-ethanimidamide dihydrochloride (1400W dihydrochloride; 10^{-4}, 10^{-3}, 10^{-2} nmol/0.2µL), or the NF-kβ inhibitor, 1-Pyrrolidinecarbodithioic acid ammonium salt (PDTC; 10^{-3}, 10^{-2}, 10^{-1} e 1nmmol/0.2 µl). Naïve or guide cannulae-implanted rats were submitted to an acute restraint stress (RS; 2h) and were maintained single-housed for additional 24h until behavioral analysis. Non-stressed and stressed rats received drugs intra-MPFC and 10 min later were tested in the elevated plus-maze (EPM) for 5 min. Results: RS induced anxiogenic-like effect and pre-treatment with 1400W (10^{-3} and 10^{-2} nmol) or PDTC (10^{-3} and 10^{-2} nmol) prevented this effect (stress and treatment effect $p<0.05$). Most importantly, the drugs did not change behavior in non-stressed rats ($p>0.05$), indicating that its effect depends on alterations induced by the stress. Conclusions: Our results showing that RS-induced anxiogenic behavior involves iNOS and NF-kβ in the MPFC suggest that targeting inflammatory mechanisms in the MPFC could treat stress-related disorders.

Disclosures: S.F. Lisboa: None. **C. Vila-Verde:** None. **D.L. Uliana:** None. **L.A. Braga:** None. **L.B.M. Resstel:** None. **F.S. Guimaraes:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.05/PP1

Topic: F.04. Stress and the Brain

Support: CONACyT 243419

Title: Neonatal administration of prolactin decreases glial population and attenuates stress-induced cytokine expression in the hippocampus of male rat pups

Authors: *L. TORNER[1], G. ZINZUN-IXTA[1], A. OCHOA-ZARZOSA[2]
[1]Inst. Mexicano Del Seguro Social, Morelia, Mexico; [2]Ctr. Multidisciplinario de Estudios en Biotecnología, Univ. Michoacana de San Nicolás de Hidalgo, Morelia, Mexico

Abstract: Abnormal activation of hormones and neurotransmitter systems early in life leads to a high vulnerability to develop physiologic alterations and produce psychiatric illness. Neonatal administration of prolactin (PRL) to rat pups was shown to reduce hippocampal neurogenesis by postnatal day (PN) 15. Since PRL is a cytokine, it probably exerts its actions through the modulation of the neuroendocrine system. Here we analyzed the effects of neonatal PRL on the morphology of hipocampal glial cells and on its cytokine expression (under basal and stress-stimulated conditions) in the hippocampus and in the periphery at PN15.
Sprague Dawley male rat pups were used. PRL (13 mg/kg bw) or vehicle were daily injected to pups through PN1-14, and other pups were left undisturbed (control). Pups were sacrificed on PN15, either under basal (no-stress) conditions or were subjected to a stressor (3h maternal separation) before sacrifice. Brains (basal) or hippocampi (basal/stress) were isolated and trunk blood was collected. Immunocytochemistry for GFAP and Iba1 was performed to analyze glial cells; qPCR was used to assess TNF-α, IL-1$_\beta$, e IL-6 expression in the hippocampus and ELISA techniques to evaluate plasma cytokine concentrations.
We show that neonatal PRL administration has no effect on microglial cell density in the hippocampus, and no change in the amount of the activated cells was found. In contrast, PRL induces a significant decrease in the astrocyte population in the hipocampal hilus. Hippocampal expression of TNF-α, IL-1$_\beta$, e IL-6 was not altered by PRL under basal conditions, but it became attenuated in response to stress. Peripheral concentrations of TNF-α increased in PRL-treated pups under basal and stressed conditions, IL-1$_\beta$ concentration decreased in PRL-treated pups after stress, and IL-6 concentration was not affected. We conclude that a chronic neonatal administration of PRL induces an attenuated response of cytokine expression in response to stress, perhaps due to a decreased population of astrocytes. (Support: grant from CONACyT 243419). No conflicts of interest are perceived.

Disclosures: L. Torner: None. **G. Zinzun-Ixta:** None. **A. Ochoa-Zarzosa:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.06/PP2

Topic: F.04. Stress and the Brain

Support: NIH Grant MH084970 and MH109484 to JAR

Title: Effects of chronic low-grade inflammation on basolateral amygdala function in rats

Authors: *S. MUNSHI, J. A. ROSENKRANZ
Rosalind Franklin Univ. of Med. & Sci., North Chicago, IL

Abstract: Systemic inflammation and infection can cause changes in mood or affect accompanied by a group of physical manifestations including lethargy, malaise, anhedonia, listlessness, decreased appetite and fever. These changes in the neurophysiologic and behavioral aspects seen with peripheral inflammation are mediated at the molecular level by proinflammatory cytokines such as interleukin (IL)-1β, IL-6, and TNF-α. The basolateral amygdala (BLA) is a key brain region involved in emotion. Recent evidence suggests that acute peripheral immune challenge by the proinflammatory cytokine IL-1β in a dose that causes sickness-like behavior alters *in vivo* BLA neuronal firing in rats. Although acute infection and inflammation are associated with the pathogenesis of sickness behavior, very little is known about how chronic, low ("subthreshold") dose of inflammation affects mood and behavior, especially at the level of the BLA neuronal involvement. In the present study, adult male Sprague Dawley rats were treated with different doses of the inflammagen IL-1β (0, 0.25, 0.50 and 1 μg, intraperitoneal (i.p.), single dose) to determine their time-dependent effects (30 min, 2h, 4h and 24h) on locomotion in the open field test. Low dose of IL-1β (0.25 μg) did not cause any significant decrease in the distance traveled and the speed of movement. Hence this dose was used to induce a chronic low-grade inflammation without causing overt sickness. We hypothesized that chronic low-grade inflammation affects BLA-dependent behaviors and increases the BLA neuronal activity. To test this hypothesis, low-dose IL-1β (0.25 μg, i.p.) was administered twice/day for five consecutive days. On the sixth day, anxiety-like behavior was evaluated by open field test, and spontaneous BLA neuronal firing rate was determined using single-unit *in vivo* extracellular electrophysiological recordings. Our preliminary behavioral data show that the chronic low-dose of IL-1β (CLD $_{(IL\text{-}1\beta)}$) causes a trend of decrease in the time spent and the distance traveled in the central area of the open field, without affecting the total distance traveled and the speed of movement, indicating that the CLD $_{(IL\text{-}1\beta)}$ causes anxiety-like behavior. Additionally, our electrophysiological data show that the spontaneous neuronal firing rate is significantly increased in the CLD $_{(IL\text{-}1\beta)}$ group compared to the control. These results indicate that chronic subthreshold inflammation can induce anxiety behavior and amygdala hyperactivity even in the absence of overt sickness. This may contribute to effects of inflammation in mood disorders.

Disclosures: S. Munshi: None. **J.A. Rosenkranz:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.07/PP3

Topic: F.04. Stress and the Brain

Title: Stress impairs memory and activates caspase-1 via a mechanism involving adrenergic signaling and adenosine

Authors: *A. E. TOWERS[1], M. L. OELSCHLAGER[2], M. R. LORENZ[3], G. G. FREUND[4]
[1]Div. of Nutritional Sci., [2]Animal Sci., [3]Program in integrative immunology and behavior, [4]Dept. of Pathology, Univ. of Illinois At Urbana-Champaign, Urbana, IL

Abstract: Inflammation is a central component to the etiology of many cognitive and mood disorders (Dantzer et al., 2008). In the brain, one of the main pro-inflammatory messengers is the cytokine IL-1β. Recent evidence suggests that stress induced by a variety of common events, such as handling, environmental changes, and metabolic disruptions can activate the IL-1 arm of the neuroimmune system leading to cognitive impairment and anxiety-like behaviors (Goshen and Yirmiya, 2009). However, the precise mechanisms by which stress activates IL-1β in the brain remain poorly understood. Here we subjected 10-week old male C57BL/6 mice to handling stress utilizing common experimental procedures (ie. weighing, scruffing, and needle prick) over a period of 6 hrs. 6 hrs of stress increased the IL-1β activating enzyme caspase-1 by 120% in the prefrontal cortex, amygdala, and hippocampus. Stress also impairs recognition memory in the novel object recognition (NOR) task by 12%. These effects of stress on NOR are mechanistically linked to IL-1β activation, as genetic deletion of both caspase-1 and IL-1 receptor 1 (IL1R1) increased NOR by 13% and 21%, respectively, as well as pharmacological inhibition of IL1R1 by 12%. To determine how the stress response hormones affect stress-induced memory impairment we administered mice propranolol and mifepristone prior to initiating stress. Propranolol increased NOR by 13%, while mifepristone had no effect on NOR suggesting that the adrenergic stress hormones mediates memory impairment due to stress. As adrenergic signaling can increase extracellular adenosine levels (Crosson and Petrovich, 1999), and we have previously shown exogenous adenosine is capable of inducing anxiety-like behaviors (Chui et al., 2014), we wanted to determine if adenosine plays a role in stress-induced memory impairment. We found that blocking adenosine signaling by administration of caffeine improved novel object performance following stress by 16%. Furthermore, the effects of adenosine during stress appear to be specific to the A2A receptor, as A2A specific antagonists, but not A1A antagonists, improved memory by 18% following stress. Inhibition of the A2A receptor also decreased caspase-1 activation by 80% in the amygdala following stress. Taken together, these data show that it is the adrenergic arm of the stress response, through its effects on adenosine signaling, that leads to IL-1β activation in the brain and cognitive impairment. These results further our understanding of how stress interacts with the neuroimmune system, as well as identify new potential targets for therapeutic intervention.

Disclosures: A.E. Towers: None. **M.L. Oelschlager:** None. **M.R. Lorenz:** None. **G.G. Freund:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.08/PP4

Topic: F.04. Stress and the Brain

Support: NIMH T32MH103213

Title: Estradiol mediates chronic stress effects on microglial morphology in medial prefrontal cortex in female rats

Authors: *J. L. BOLLINGER, C. L. WELLMAN
Psychological and Brain Sci., Indiana Univ., Bloomington, IN

Abstract: Recent studies indicate a role for neuroimmune pathways and microglia in psychological health and disease. Microglia can regulate synaptic plasticity by releasing neuroactive factors, directly pruning dendritic spines, and modulating neuronal architecture. We have shown that stress differentially affects microglial cell activation and immune factor expression in males and females in a number of brain regions critical for emotion regulation and cognitive function, including medial prefrontal cortex (mPFC). Contrary to findings in male rats, chronic stress reduces microglial morphological activation state in the prelimbic region (PL) of mPFC in females. To investigate the potential role of 17β-estradiol (E) in stress-induced microglial remodeling in female rats, animals underwent sham surgeries (SHAM), ovariectomies (OVX), or ovariectomies with E replacement (OVX+E). After a recovery period, rats received daily restraint stress (3 h/day, 10 days) or were left unhandled except for weighing. Following the final day of restraint, brains were processed for visualization of microglia via Iba-1 immunohistochemistry. In 4 sections evenly spaced through the rostral-caudal extent of PL, the total area of Iba-1-positive (Iba-1+) material was assessed within layers 1, 2/3, 5, and 6 (for a total of 32 separate 67,500 μm^2 sampling areas per rat) using a semi-automated thresholding technique. The total area of Iba-1+ material was similar across unstressed groups regardless of hormonal manipulation. Congruent with previous findings, chronic stress reduced the total area of Iba-1+ material in mPFC in SHAM females, suggesting a reduction in morphological activation state. Chronic stress had no effect on the total area of Iba-1+ material in OVX females. However, the chronic stress-induced reduction in Iba-1+ material was restored in OVX+E animals. Thus, estradiol mediates the effects of chronic stress on microglial morphology in mPFC in female rats. This suggests that gonadal hormones may contribute to the differential effects of stress on microglial biology in mPFC and, in turn, neural structure, function, and behavior in males versus females.

Disclosures: J.L. Bollinger: None. **C.L. Wellman:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.09/PP5

Topic: F.04. Stress and the Brain

Support: NIH Grant R01MH108523

Title: Exposure to an acute stressor disinhibits microglia via down-regulation of CD200R: A mechanism of neuroinflammatory and microglial priming

Authors: *M. G. FRANK, *M. G. FRANK, L. K. FONKEN, J. ANNIS, L. R. WATKINS, S. F. MAIER
Univ. of Colorado, Boulder, CO

Abstract: Prior exposure to acute and chronic stressors has been found to potentiate the neuroinflammatory and microglial response to subsequent immune challenges, suggesting that stress shifts the activation state of microglia from surveillant to primed. Interestingly, a primed immunophenotype has also been observed in animals lacking CD200, a neuronal and endothelial membrane glycoprotein that inhibits microglia through its cognate receptor CD200R. Thus, we explored the possibility that disruption of CD200:CD200R signaling may be involved in stress-induced neuroinflammatory priming. Male Sprague-Dawley rats were exposed to inescapable tailshock (IS) or served as home cage controls (HCC), and 3 separate experiments were conducted to measure mRNA expression of CD200R 24 hr after stress in 1) whole hippocampus, 2) micropunches of hippocampal sub-regions (CA1, CA3 and dentate gyrus) and 3) hippocampal microglia. We found that IS induced a consistent and robust down-regulation of CD200R in each experiment. Additional immunophenotypic markers (MHCII, TLR4, RAGE, CX3CR1 and CD200) as well as pro-inflammatory mediators (IL-1 beta, IL-6, TNFa, NF-kBIA and NLRP3) were measured in these experiments, but only CD200R was significantly altered by stress at the 24 hr timepoint. This down-regulation of CD200R suggested that CD200:CD200R inhibitory signaling of microglia may similarly be down-regulated, thereby inducing a primed immunophenotype in microglia. To test this notion, male Sprague-Dawley rats were injected intra-cisterna magna with a soluble fragment of CD200 (CD200 Fc; 5 ug) or a control protein (human IgG1 Fc) in sterile PBS. Immediately after injection, animals were exposed to IS or served as HCC. 24 hr after IS treatment, hippocampal microglia were isolated, suspended in media (DMEM + 10% FBS) and treated with the TLR4 agonist lipopolysaccharide (LPS; 0, 1, 10 and 100 ng/ml) for 2hr. mRNA of pro-inflammatory mediators was then measured using real time RT-PCR. Consistent with prior findings, exposure to IS potentiated the pro-inflammatory response (IL-1 beta, TNFa, NF-kBIA, and NLRP3) to LPS compared to the cytokine response in HCC. Treatment with CD200 Fc blocked this stress-induced potentiation of the pro-inflammatory response to levels observed in HCC animals, suggesting that CD200 Fc had abrogated stress-induced priming of microglia by increasing inhibitory drive via CD200R. Taken together, the present findings suggest that stress-induced down-regulation of CD200R on microglia may serve to disinhibit microglia, thereby priming the microglial pro-inflammatory response to subsequent immune challenges.

Disclosures: **M.G. Frank:** None. **L.K. Fonken:** None. **J. Annis:** None. **L.R. Watkins:** None. **S.F. Maier:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.10/PP6

Topic: F.04. Stress and the Brain

Support: H2020 Marie-Curie IF 20771

Title: Neuromolecular mediators of resilience to chronic stress in the bed nucleus of the stria terminalis of the mouse

Authors: *A. GURURAJAN[1], J. M. LYTE[2], A. VENTURA DA SILVA[2], G. M. MOLONEY[1], T. M. BECKER[2], R. O. CONNOR[2], M. BOEHME[2], M. V. WOUW[2], *B. MERCX[1], T. G. DINAN[2], J. F. CRYAN[1]
[1]Anat. & Neurosci., [2]APC Microbiome Inst., Univ. Col. Cork, Cork, Ireland

Abstract: Chronic stress is a major risk factor for the onset of various psychopathologies. However, not all who are exposed go on to develop the symptomatology that is associated with these disorders, a phenomenon termed stress resilience. Several key limbic structures have been implicated as regulators of the stress response, namely the prefrontal cortex, amygdala and the hippocampus. Their effects on the HPA axis are via subthalamic relay structures such as the bed nucleus stria terminalis (BNST). Thus, the BNST is centrally placed to gate the response to stress and may play an unexplored, critical role in determining whether an individual is stress resilient or stress susceptible. The aim of our study was to identify correlates of stress resilience and stress susceptibility. Additionally, we wanted to characterise the neuromolecular phenotypes of the BNST in both groups of mice. Male C57Bl/6 mice were subjected to the chronic psychosocial defeat stress paradigm after which they were tested for social avoidance behaviour and labelled as either stress-resilient or stress-susceptible based on test performance. Prior to the start of the paradigm and the day after the last defeat session, tail blood was collected for analysis of plasma corticosterone levels. The day after behavioural testing mice were culled. Trunk blood was collected and used to characterise the immunophenotype by flow cytometry. Brains were dissected out into several brain regions which were processed to analyse expression of genes implicated in the stress-response; for some mice, the hippocampus was used for microelectrode array experiments. Peripheral corticosterone levels were higher in stress susceptible than stress resilient mice before and after the social defeat stress paradigm. PCR analyses revealed regionally specific effects on the expression of corticotrophic release factor and its receptors, gluco- and mineralocorticoid receptors and FKBP5. RNA-Seq of the BNST is ongoing to identify differential expression of coding and non-coding genes. Flow cytometry and electrophysiology analyses are also underway to detect differences between stress resilient and stress susceptible mice. **Conclusion:** Consistent with recent literature, we have shown that stress

resilience is defined by a complex molecular and physiological profile which spans central and peripheral compartments. Stress resilience could be predicted based on baseline peripheral corticosterone levels. Importantly, our research will potentially reveal a role for the BNST not just as a relay station for limbic input but also a structure involved in neuroadaptative processes that facilitate the development of stress resilience.

Disclosures: **A. Gururajan:** None. **J.M. Lyte:** None. **A. Ventura Da Silva:** None. **G.M. Moloney:** None. **T.M. Becker:** None. **R.O. Connor:** None. **M. Boehme:** None. **M.V. Wouw:** None. **B. Mercx:** None. **T.G. Dinan:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Mead Johnson, Cremo, 4D Pharma, Suntory Wellness, Nutricia. **J.F. Cryan:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Mead Johnson, Cremo, 4D Pharma, Suntory Wellness, Nutricia.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.11/PP7

Topic: F.04. Stress and the Brain

Support: AFOSR-2016-0018A

Title: Acute-stress induced gastrointestinal serotonergic responses are region-dependent and host strain specific

Authors: *J. M. LYTE[1], M. S. GOODSON[3], N. KELLEY-LOUGHNANE[3], T. G. DINAN[2], J. F. CRYAN[2], G. CLARKE[2]
[2]APC Microbiome Inst., [1]Univ. Col. Cork, Cork, Ireland; [3]711th Human Performance Wing, Air Force Res. Lab. Wright-Patterson Air Force Base, Dayton, OH

Abstract: Objective: Host genetics influence the acute stress response, the impact of which is frequently manifested in altered gastrointestinal function and exacerbated in brain-gut axis disorders such as irritable bowel syndrome.Gut-derived serotonin (5-HT) produced following a stressor can exert physiologically and clinically important local and systemic effects. We sought to define the gastrointestinal serotonergic system response to and recovery from an acute stressor in genetically-distinct mice strains. **Methods:** Adult male NIH Swiss Webster, BALB/c, and C57/BL6 mice were randomly allocated to the unstressed control or stress group. Stressed animals were subjected to 15min of restraint stress (n=4-8 mice/timepoint/strain) and sacrificed

post-stressor +0, 5, 15, 30, 45, 60, or 240min. Plasma corticosterone was assayed using an ELISA. Ileal and colonic 5-HT and 5-HIAA concentrations were determined using HPLC. Results were analyzed by student's t-test or ANOVA, where applicable, and statistical significance was set at a $p<0.05$. **Results:** Plasma corticosterone was significantly ($p<0.05$) elevated immediately after restraint stress compared to control group in each strain. C57/BL6 exhibited a greater ($p<0.05$) plasma corticosterone concentration post-stressor compared to BALB/c or NIH Swiss Webster mice. Colonic 5-HT levels were higher than ileal 5-HT in all mouse strains. Strain differences in distal ileal 5-HT and 5-HIAA concentrations at baseline were maintained post-stressor. Ileal 5-HT and 5-HIAA concentrations were frequently highest in C57/BL6 compared to other strains at baseline and post-stress. Proximal colonic 5-HT and 5-HIAA were different ($p<0.05$) at several post-stressor timepoints in C57/BL6 compared to other strains. Analyses are ongoing for neural correlates following an acute stressor. **Conclusions:** Host genetics heavily influences the stress response. The C57/BL6 strain displayed the largest post-stress HPA axis activity and had higher levels of 5-HT in the colon and ileum at multiple timepoints post-stressor. Further studies are required to understand the implications of these findings for the control of stress-induced 5-HT-mediated gastrointestinal symptoms and to assess the role of the gastrointestinal microbiota and microbial metabolites in regulating the local gastrointestinal serotonergic system response to acute stressors.

Disclosures: J.M. Lyte: None. **M.S. Goodson:** None. **N. Kelley-Loughnane:** None. **T.G. Dinan:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Cremo, 4D Pharma, Suntory Wellness, Mead Johnson, Nutricia. **J.F. Cryan:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Cremo, 4D Pharma, Suntory Wellness, Mead Johnson, Nutricia. **G. Clarke:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.12/PP8

Topic: F.04. Stress and the Brain

Title: The effects of stress-inflammation interaction on behavior and gene expression in mice models of mood and anxiety disorders

Authors: *S. FLAISHER-GRINBERG[1], L. OLEK[2], J. KUEHN[2]
[1]Dept. of Psychology, [2]St. Francis Univ., Loretto, PA

Abstract: Chronic exposure to stress affects a variety of physiological systems and has been linked to the activation or amplification of pathological conditions such as depression, Post-Traumatic Stress Disorder and Alzheimer's. While neuroinflammation has been implicated in the deleterious outcomes of chronic stress, the effects of the timing, and regimen of immune activation on brain area-specific gene expression and behavioral functioning is yet to be determined. The current project evaluated the effects of acute and repeated Lipopolysaccharide (LPS, a pro-inflammatory agent) administration, before or during the exposure of C57BL6/J mice to the unpredictable chronic mild stress paradigm. Behavioral effects were assessed using the Open Field, Black-White box, Elevated-Plus Maze, Tail-Suspension Test and Forced Swim Test, and the expression of NPY1R, NPYR2, BDNF and HTR2C genes was evaluated in the hippocampus, hypothalamus, amygdala and medial-prefrontal cortex of C57BL6/J mice using qPCR. Results demonstrate that the combination of chronic stress exposure and pro-inflammatory challenge yield differential effects on mood and anxiety-related behaviors, as well as region-specific gene expression. The findings provide insight into the mechanisms regulating the behavioral response to stress and inflammation, and highlight particular targets for brain area-specific intervention.

Disclosures: S. Flaisher-Grinberg: None. **L. Olek:** None. **J. Kuehn:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.13/PP9

Topic: F.04. Stress and the Brain

Support: Milgrom Family Foundation

NATO SPS Programme

Herman Dana Foundation

Title: Peripheral blood gene expression signature accompanying acute stress exposure among adolescents with and without early childhood adversity

Authors: *T. GOLTSER[1,2,3], C. KALLA, 7174166[2], A. SHALEV[3], F. BEN-HARASH[3], R. GIESSER[3], A. BEN YEHUDA[4], O. OZ[3], D. PEVZNER[3], I. VASHDI[3], R. HABER[2,3], C. SALONER[2,3], A. MIRAN[2], L. CANETTI[2], E. GALILI-WEISSTUB[3], O. BONNE[2], R. SEGMAN[2]
[1]Psychiatry, Kiryat Hadassah, Jerusalem, Israel; [2]Mol. Psychiatry Lab. - Dept. of Psychiatry, Hadassah - Hebrew Univ. Med. center, Jerusalem, Israel., Jerusalem, Israel; [3]The Herman-Danna

Div. of Pediatric Psychiatry, Dept. of Psychiatry, Hadassah - Hebrew Univ. Med. Center;, Jerusalem, Israel; [4]Dept. of Mental Hlth. Israel Def. Forces., Jerusalem, Israel

Abstract: Background: Early childhood adversity has been previously implicated with lifelong altered stress reactivity. Maladaptive stress reactivity may result in compromised neurocognitive and emotional regulation aptitudes and may lead to acute and chronic post traumatic stress disorder and depression. The current study explores the predictive value of blood based expression changes for identifying individual stress related vulnerability. **Methods:** Several hundred adolescent trainees undergoing combat military training were screened for exposure to early childhood trauma. An extremes case control sample compared those with high and low childhood trauma for prospective anxiety depressive and post traumatic symptoms, neuropsychological measures, and blood sampling, at rest and during exposure to extreme stress under simulated combat training conditions. Premorbid characteristics and neuropsychological and biological measures were employed to predict mal/adaptive outcomes following simulated combat. **Results:** Distinct blood expression profiles and psychological measures mark stress vulnerability and may help predict longer term outcomes before and immediately following exposure to trauma. **Conclusion:** Distinct signatures in blood may help focus preventive and interceptive efforts on vulnerable subjects during stress exposure and at its immediate aftermath, before chronic PTSD or depression develop. Results may further shed light on the role of inflammatory and neuro endocrine reactivity in mediating the transduction of stress into long term maladaptive neuropsychiatric outcomes.

Disclosures: T. Goltser: None. **C. Kalla:** None. **A. Shalev:** None. **F. Ben-Harash:** None. **R. Giesser:** None. **A. Ben Yehuda:** None. **O. Oz:** None. **D. Pevzner:** None. **I. Vashdi:** None. **R. Haber:** None. **C. Saloner:** None. **A. Miran:** None. **L. Canetti:** None. **E. Galili-Weisstub:** None. **O. Bonne:** None. **R. Segman:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.14/PP10

Topic: F.04. Stress and the Brain

Support: Israel Science Foundation Grant 1563-08

the Herman Dana Foundation

Title: Mononuclear cell gene expression changes in postpartum depression

Authors: *R. SEGMAN[1,2,3], T. GOLTSER DUBNER[2,3], T. SHIMONOVITZ[4], S. KLAR[2], L. CANETTI[2], E. GALILI-WEISSTUB[3], I. SHACHAR[2], D. PEVZNER[2,3], O. OZ[2,3], I. VASHDI[2,3],

N. FRIEDMAN[5], D. HOCHNER-CELNIKIER[4]
[1]Hadassah Univ. Hosp., Jerusalem, Israel; [2]Mol. Psychiatry Lab. Dept. of Psychiatry, Hadassah - Hebrew Univ. Med. center,, Jerusalem, Israel; [3]The Herman-Danna Div. of Pediatric Psychiatry, Dept. of Psychiatry, Hadassah - Hebrew Univ. Med. Center;, Jerusalem, Israel; [4]Dept. of Obstetrics and Gynecology - Hadassah Mt. Scopus - Hebrew Univ. Med. center, Jerusalem, Israel; [5]Sch. of Computer Sci. and Engineering, Hebrew Univ., Jerusalem, Israel

Abstract: Background: Altered immune cell reactivity during the development of depression after delivery, may point to biomarkers and potentially implicate underlying immune mechanisms. Methods: Genome scale mononuclear expression patterns after delivery were compared between mothers prospectively diagnosed with depression and resilient mothers. Results: Differential immune cell gene expression patterns associate significantly enriched pathways revealing altered immune function that accompany the triggering of postpartum depression. Pathway involvement implicate relevant immune candidates. Discussion: Differential mononuclear cell transcripts sampled postpartum point to unique immune activation among depressed mothers. Beyond serving as potential biomarkers, findings may point to pathogenetically relevant molecular targets involved in the development of depression.

Disclosures: R. Segman: None. **T. Goltser Dubner:** None. **T. Shimonovitz:** None. **S. Klar:** None. **L. Canetti:** None. **E. Galili-Weisstub:** None. **I. Shachar:** None. **D. Pevzner:** None. **O. Oz:** None. **I. Vashdi:** None. **N. Friedman:** None. **D. Hochner-Celnikier:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.15/PP11

Topic: F.04. Stress and the Brain

Title: Effects of repeated stress on the ying-yang of catecholamines and glucocorticoids in the regulation of brain cytokines

Authors: *D. F. BARNARD[1], K. GABELLA[1], A. KULP[2], A. PARKER[1], J. D. JOHNSON[3]
[2]Biomed. Sci., [3]Biol Sci. Dept, [1]Kent State Univ., Kent, OH

Abstract: The elevation in pro-inflammatory cytokines in the brain, particularly IL-1β, following stress exposure have potent behavioral and physiological altering effects. Stress-induced cytokine responses are tightly regulated by catecholamines and glucocorticoids, which have opposing regulatory control over pro-inflammatory cytokine production. Specifically, catecholamines enhance cytokine production via beta-adrenergic receptors (β-ARs) while glucocorticoids (largely corticosterone in rodents) suppress cytokine production. The aim of this current study was to examine how repeated stress exposure effects the regulation of brain pro-

inflammatory cytokines. Fischer rats were exposed to four days of repeated mild stress and 24h following the last stressors animals were administered vehicle, propranolol (β-AR antagonist), metyrapone (corticosterone synthesis inhibitor) or combination of propranolol and metyrapone. Two hours after drug injections, limbic areas (Amygdala, Hippocampus, and Hypothalamus) were collected and processed for measurement of IL-1β mRNA. Stress had no effect on basal IL-1β mRNA levels in vehicle treated animals. Administration of propranolol had no effect in non-stressed controls; however, it did significantly decrease IL-1β mRNA in stress animals in the amygdala and hippocampus. Metyrapone significantly increased IL-1β mRNA in both control and stressed animals in the amygdala and hypothalamus, but the increase was blocked by prior administration of propranolol. These data indicate that β-AR stimulation is critical for maintaining basal IL-1β production in the amygdala and hippocampus of animals exposed to repeated stress. Furthermore, the increase in levels of IL-1β mRNA following blockade of corticosterone synthesis is dependent on β-AR stimulation, suggesting glucocorticoids may regulate brain cytokines via modulating norepinephrine release. Currently, similar studies using female animals will uncover potential sex differences in the effect repeated stress has on the regulation of brain IL-1β production.

Disclosures: D.F. Barnard: None. **K. Gabella:** None. **A. Kulp:** None. **A. Parker:** None. **J.D. Johnson:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.16/PP12

Topic: F.04. Stress and the Brain

Support: Susquehanna University Scholarship Mini-Grant

Title: Sex-differences in prenatal stress model of microglial activation in stress-regulating structures

Authors: *S. N. CASSELLA, E. J. JONES, D. WILSON, A. GRANT, E. RHINEHART
Susquehanna Univ., Selinsgrove, PA

Abstract: Prenatal stress has shown to have adverse effects on various aspects of the developing brain, such as alterations in neuroendocrine functioning and neuroinflammatory processes. These changes may directly contribute to the development of psychiatric disorders such as addiction, anxiety, and schizophrenia. Psychiatric disease research has found an association between these disorders and altered immune function. The innate immune cells of the central nervous system are microglia, and they range in morphology from ramified with a surveying function, to amoeboid with a pro-inflammatory function. Neuroinflammation occurs when microglia

accumulate and become active. We sought to determine if our prenatal stress model would induce neuroinflammation that would correlate with previously reported changes in neuroendocrine and behavioral outcomes, and further, if these alterations differed between the sexes. To model prenatal stress, we used a food deprivation paradigm (FD50) in which food was restricted by 50% during the second half of pregnancy. We examined microglia in the stress response-regulating regions of the hippocampus, prefrontal cortex (PFC), amygdala, and hypothalamus. Offspring brains were collected at postnatal day (P) 30 and P60 and microglial morphology was analyzed via soma area and branching complexity to determine differences in relative activation between groups. Ongoing analysis shows a significant increase in soma size between the control and restricted groups in only the infralimbic PFC of females, suggesting that these cells are functioning in a relatively activated state. Understanding the relationship between prenatal stress, inflammation and behavioral alterations could lead to enhanced prenatal care and therapies in the future.

Disclosures: S.N. Cassella: None. **E.J. Jones:** None. **D. Wilson:** None. **A. Grant:** None. **E. Rhinehart:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.17/PP13

Topic: F.04. Stress and the Brain

Support: NARSAD Young Investigator Award

Title: Microglia during early life contribute to myelination, HPA axis development, and the behavioral effects of neonatal stress

Authors: *L. H. NELSON[1], S. WARDEN[2], K. M. LENZ[3]
[1]Ohio State Univ., Columbus, OH; [2]Dept. of Neurosci., The Ohio State Univ., Columbus, OH; [3]Psychology, Ohio State Univ. Dept. of Psychology, Columbus, OH

Abstract: Microglia, the brain's resident immune cells, regulate many processes of normal brain development, including cell genesis, cell death, myelination, axon guidance, and synaptic patterning (Pierre et al., Brain Behav Immun 2016). Early life perturbations, such as stress, perturb microglial function, and increase anxiety-like behavior (Delpech et al., Brain Behav Immun, 2016). We have previously shown that neonatal microglial depletion during the early postnatal period decreases anxiety, behavioral despair and the acute stress response in adulthood (Nelson et al., 2016, Bev Brain Res). Yet the mechanisms through which microglia impact the early life programming of mood related behavior at baseline or in response to early life perturbations remains unknown. To probe the mechanisms through which microglia contribute to

such early life programming of behavior, we centrally infused liposomal clodronate (2μL icv; Encapsula Nanoscience) on postnatal days (P) 1 and 4, which temporarily depletes 90% of forebrain microglia. To determine the brain regions responsible for the dampened HPA axis response we previously observed in microglia depleted animals, we assessed cFos expression in the medial prefrontal cortex (mPFC) after acute restraint stress in adults. We found decreased cFos expression in the mPFC in clodronate treated animals relative to controls. We are currently assessing the cFos expression in the amgydala to determine if there is region-specific sensitivity to early life programming effects of microglia. Several studies have found changes in synaptic- and myelination-related gene expression due to early life stress (Wei et al., 2015 Dev Neuro; Bath et al., 2016 Horm Bev) thus microglia may be programming mood-related behavior via underlying changes in these endpoints. We assayed the gene expression of several synaptic genes (NR2A, NR2B, PSD95, vGlut1) and the developmental time-course of myelin basic protein (MBP) in the mPFC and amygdala following microglial depletion. We found no changes in synaptic genes at P12 in the amygdala. However, we found that MBP was decreased at P12 in the mPFC and amygdala, and still decreased at P22 in the amygdala but increased at P22 in the mPFC. Currently, we are combining early life microglial depletion with maternal separation stress to determine whether microglia are necessary for stress-induced programming of mood-related behavior and underlying perturbations in brain development. These studies are important for elucidating the functional role that microglia play in normal and abnormal development of the brain and behavior.

Disclosures: L.H. Nelson: None. **S. Warden:** None. **K.M. Lenz:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.18/PP14

Topic: F.04. Stress and the Brain

Support: PROPe-UNESP Grant 12/2015-PROPe

CAPES-DS Grant

Title: β-adrenergic signaling suppression protects against chemically induced carcinogenesis in rats

Authors: *V. B. VALENTE[1], F. VERZA[1], H. CECÍLIO[1], F. LOPES[1], M. SUNDEFELD[1], K. TJIOE[1], É. BIASOLI[1], G. MIYAHARA[1], A. SOUBHIA[1], C. FURUSE[1], M. DE ANDRADE[4], S. DE OLIVEIRA[2], D. BERNABÉ[3]
[1]Oral Oncology Center, Dept. of Pathology and Clin. Propedeutics, [2]Oral Oncology Center, Dept. of Basic Sci., [3]Psychoneuroimmunology Res. Center, Oral Oncology Ctr., São Paulo State

Univ. - UNESP, Aracatuba, Brazil; [4]Dept. of Biomed. Statistics and Informatics, Mayo Clin. Col. of Med., Rochester, MN

Abstract: β-adrenergic signaling has been found to affect important intracellular processes that contribute to cancer development and progression. In the tumor microenvironment, β-adrenergic receptors on tumor cells are activated by norepinephrine released from peripheral sympathetic nerve fibers. Clinical and pre-clinical investigations have associated the use of propranolol (a non-specific β-antagonist) to reduced rates of progression for solid tumors. Although norepinephrine-activated β-adrenergic signaling has been increasingly linked to cancer progression, this neuroendocrine mechanism in the carcinogenesis context is still poorly understood. In this pre-clinical study, we have used a new methodology based on oral carcinogenesis model in rats to test the hypothesis that norepinephrine levels in the normal tissue microenvironment pre-carcinogenic induction would have a significant predictive value for cancer occurrence and progression. Male Wistar rats were underwent to a normal tongue tissue biopsy for measuring norepinephrine levels before carcinogenic induction with 4-nitroquinoline-1-oxide (4NQO) carcinogen. Then, animals were euthanized for evaluation of oral squamous cell carcinoma (OSCC) or oral leukoplakia (non-cancerous lesion) occurrence into tongue. Increased norepinephrine concentrations in the tissue microenvironment before 4NQO treatment were predictive for OSCC occurrence. Higher pre-carcinogen hormone levels were also correlated to CDKN2a-p16 mRNA lower expression in OSCCs. Furthermore, increased norepinephrine concentrations in OSCC were associated to a higher tumor volume and thickness, and with a lesser intensity of the lymphoplasmocytic infiltrate underlying to tumor. In another experimental set, to assess the effects of adrenergic signaling on OSCC incidence, animals were treated with propranolol or only saline solution (sham group) during carcinogenic induction. Propranolol treatment inhibited OSCC incidence and reduced cancer-related cachexia. This study demonstrates that increased pre-carcinogen norepinephrine levels in the normal tissue microenvironment can be predictive for chemically induced carcinogenesis in rats. Moreover, the suppression of adrenergic signaling protected these animals from the cancer development.

Disclosures: V.B. Valente: None. **F. Verza:** None. **H. Cecílio:** None. **F. Lopes:** None. **M. Sundefeld:** None. **K. Tjioe:** None. **É. Biasoli:** None. **G. Miyahara:** None. **A. Soubhia:** None. **C. Furuse:** None. **M. de Andrade:** None. **S. de Oliveira:** None. **D. Bernabé:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.19/PP15

Topic: F.04. Stress and the Brain

Support: Undergraduate Research Opportunity Program (UROP) Assistantships to AKL, TMS, EMS, AAO, and KSH

UROP Individual Grants to TSM, and CJB

UROP/Howard Hughes Medical Institute (HHMI) Individual Awards to AKL and CJB

Biological Sciences Initiative Biosciences Undergraduate Research Skills and Training Program (BURST) awards to AKL and CJB

Title: Preimmunization with heat-killed Mycobacterium vaccae enhances fear extinction in the rat fear-potentiated startle paradigm

Authors: *J. E. HASSELL, JR[1], J. H. FOX[1], P. H. SIEBLER[1], M. R. ARNOLD[1], A. K. LAMB[1], D. G. SMITH[2], H. E. W. DAY[3], T. SMITH[1], E. M. SIMMERMAN[1], A. A. OUTZEN[1], K. S. HOLMES[1], C. J. BRAZELL[1], C. A. LOWRY[1]
[1]Integrative Physiol., [2]Chem. and Biochem., [3]Psychology and Neurosci., Univ. of Colorado, Boulder, CO

Abstract: The hygiene hypothesis or "Old Friends" hypotheses proposes that inflammatory diseases are increasing in modern urban societies, due in part to reduced exposure to microorganisms that drive immunoregulatory circuits, and a failure to terminate inappropriate inflammatory responses. Inappropriate inflammation is also emerging as a risk factor for trauma-related, anxiety, and affective disorders, including posttraumatic stress disorder (PTSD), which is characterized as persistent reexperiencing of the trauma after a traumatic experience. Traumatic experiences can lead to long lasting fear memories and fear potentation of the acoustic startle reflex. The acoustic startle reflex is an ethologically relevant reflex and can be potentiated in both humans and rats through Pavlovian conditioning. *Mycobacterium vacca*e is a soil-derived, bacterium with immunoregulatory and anti-inflammatory properties that has been demonstrated to confer stress resilience in mice. Here we immunized adult male Sprague Dawley rats 3x, once per week, with a heat-killed preparation of *M. vaccae* NCTC 11659 (0.1 mg, s.c., in 100 ul borate-buffered saline) or vehicle, and, then, 3 weeks following the final immunization, tested them in the fear-potentiated startle paradigm; controls were maintained under home cage control conditions throughout the experiment (n = 11-12 per group). Rats were tested on days 1 and 2 for baseline acoustic startle, received fear conditioning on days 3 and 4, and underwent fear extinction training on days 5-10. Rats were euthanized on day 11 and brain tissue was sectioned for analysis of *tph2, htr1a, slc6a4*, and *slc22a3* mRNA expression throughout the brainstem dorsal and median raphe nuclei. Immunization with *M. vaccae* had no effect on baseline acoustic startle or fear expression on day 5. However, *M. vaccae*-immunized rats showed enhanced between-session and with-session extinction on day 6, relative to vehicle-immunized controls. Immunization with *M. vaccae* and fear-potentiated startle altered serotonergic gene expression in a gene- and subregion-specific manner. These data are consistent with the hypothesis that immunoregulatory strategies, such as preimmunization with *M. vaccae*, have potential for prevention of stress- and trauma-related psychiatric disorders.

Disclosures: **J.E. Hassell:** None. **J.H. Fox:** None. **P.H. Siebler:** None. **M.R. Arnold:** None. **A.K. Lamb:** None. **D.G. Smith:** None. **H.E.W. Day:** None. **T. Smith:** None. **E.M. Simmerman:** None. **A.A. Outzen:** None. **K.S. Holmes:** None. **C.J. Brazell:** None. **C.A. Lowry:** F. Consulting Fees (e.g., advisory boards); CAL serves on the Scientific Advisor Board of Immodulon Therapeutics, Ltd..

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.20/PP16

Topic: F.04. Stress and the Brain

Support: FAPESP 2015/26459-5

Title: Influence of depressive-like behavior on tumor onset and progression in a chemically induced cancer model

Authors: *D. G. BERNABE, L. KOBAYASHI, F. VERZA, V. VALENTE, J. FIGUEIRA, B. SARAFIM-SILVA, M. CRIVELINI, M. SUNDEFELD, É. BIASOLI, G. MIYAHARA, S. OLIVEIRA
São Paulo State Univ., Araçatuba, Brazil

Abstract: Cancer is often accompanied by psychological disorders. Some studies have suggested that psychological or behavior traits like depression are associated with cancer incidence and progression. Head and neck cancer (HNC) patients can show higher levels of psychological disorders like depression, but the association between depression-related behavioral phenotype and susceptibility to tumor is poorly known. In this study we have used a oral carcinogenesis model to test the hypothesis that high-depressive phenotype would result in an increased tumor incidence and progression of HNC chemically induced cancer. Wistar rats were phenotyped as high- or low-depressive at baseline prior to tumor induction using forced swim test. The rats were treated with carcinogen 4-nitroquinoline 1-oxide (4NQO) during 5 months for inducing oral squamous cell carcinomas (OSCCs). Tongue sections were analyzed by microscopy for OSCC occurrence and aggressiveness diagnosis. OSCC incidence was 30% higher in the high-depressive group compared to low-depressive animals. Rats with high-depressive phenotype showed increased tumor volume than those with lower levels of depression-like behavior. Moreover, tumors from high-depressive rats had a greater degree of nuclear pleomorphism. There were no differences between the two groups regarding other clinicopathological features as weight loss, tumor thickness, pattern of tumor invasion and microscopy lymphoplasmacytic infiltrate. The depression-like behavior was also examined after chemically induced carcinogenesis and depression levels were positively correlated with tumor size. These results

suggest that high trait depression may influence susceptibility to tumor initiation and/or growth in chemically induced cancers.

Disclosures: D.G. Bernabe: None. **L. Kobayashi:** None. **F. Verza:** None. **V. Valente:** None. **J. Figueira:** None. **B. Sarafim-Silva:** None. **M. Crivelini:** None. **M. Sundefeld:** None. **É. Biasoli:** None. **G. Miyahara:** None. **S. Oliveira:** None.

Poster

413. Stress, Inflammation, and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 413.21/PP17

Topic: F.04. Stress and the Brain

Title: Microglial responses in the dentate gyrus after 72 hours sleep deprivation in mice

Authors: *C. TSAO[1], L.-H. TUAN[1], L.-J. LEE[1,2,3]
[1]Grad. Inst. of Anat. and Cell Biol., [2]Inst. of Brain and Mind Sci., [3]Neurobio. and Cognitive Sci. Ctr., Natl. Taiwan Univ., Taipei, Taiwan

Abstract: Sleep insufficiency is a serious health issue that has been linked with various physiological and psychological problems. In the present study, we adopted the multiple platform paradigm to deprive ERM sleep in adult mice. After 72 hours sleep deprivation, adult neurogenesis in the dentate gyrus was dramatically reduced. Since microglia play important roles in this process, we examined the morphometric features of microglia in the dentate gyrus. The density and morphology of Iba1-positive microglia were measured in the molecular layer, granular cell layer, subgranular zone and hilus subregions of the dentate gyrus, respectively. Compared with normal sleep and large platform controls, the density of microglia in sleep-deprived mice was increased in the hilus, while in other regions, the numbers were not altered. The morphology of microglia was also checked in a subregion-specific manner. In the molecular layer, the shape of microglia was not changed; whereas in all other subregions, the number and length of ramified microglial segments were reduced in 72-hour sleep-deprived mice. These results indicated a subregion-specific microglial responses in the dentate gyrus after sleep deprivation which could be correlated with the reduction of neurogenesis and impaired memory function in subjects of insufficient sleep.

Disclosures: C. Tsao: None. **L. Tuan:** None. **L. Lee:** None.

Poster

414. Thermoregulation and Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 414.01/PP18

Topic: F.07. Autonomic Regulation

Support: Department of Psychology Research Support Grant

Title: Yawning as a thermoregulatory response to manipulations of brain temperature

Authors: *M. L. SHOUP-KNOX, Z. FALK, M. D. KEITER
Psychology, James Madison Univ. Dept. of Psychology, Harrisonburg, VA

Abstract: The current study aims to measure brain temperature and the potentially thermoregulatory behavior of yawning in rats. Previous literature has shown that increases in brain temperature are followed by the onset of yawning, suggesting a thermoregulatory function which restores the brain to cooler temperatures (Shoup-Knox et al., 2011). Additionally, increasing environmental temperature results in increased yawning frequency as well as other thermoregulatory behaviors in parakeets (Gallup, Miller & Clark, 2009, 2010), white-faced capuchins (Campos & Fedigan, 2009, also see Gallup, 2010), and rats (Gallup, Miller & Clark, 2012). To elucidate the relationship of brain temperature and yawning frequency, independent of ambient temperature effects, it is necessary to directly manipulate brain temperature above and below the normative set point and observe the changes in yawn frequency. We propose that as we increase brain temperature we will observe an increase in yawning behavior, while decreasing the brain temperature, or holding it constant, will decrease or eliminate yawning behavior. Brain temperature was manipulated via close-ended stainless steel cannulas that were surgically implanted into the pre-optic anterior hypothalamus (POA), the area of the brain responsible for thermoregulatory behaviors. During each of four trials brain temperatures were collected and averaged over an initial 5-minute period to determine baseline. Then the POA was perfused with to water either 3° above baseline (hot condition), 1° above baseline (warm condition), or 3° below baseline (cool condition) brain temperature. Trials were each 30 minutes in duration (15 minutes water perfusion followed by 15 minutes no perfusion/recovery) and the temperature condition order was counterbalanced across animals. A control condition, also experienced by each animal, preserved baseline brain temperature. Yawning behavior was observed and recorded across the duration of each 30 minute trial. Preliminary analyses indicate that yawning occurs less frequently when baseline POA temperature is maintained and that yawning behaviors across all conditions are more likely to occur during the second 15-minute interval (recovery) of the trial. This suggests that yawning functions to restore homeostatic brain temperatures in response to warming and cooling. These data provide support for the hypothesis that yawning has evolved has as a thermoregulatory function.

Disclosures: **M.L. Shoup-Knox:** None. **Z. Falk:** None. **M.D. Keiter:** None.

Poster

414. Thermoregulation and Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 414.02/PP19

Topic: F.07. Autonomic Regulation

Support: NIH Grant RO1 AG047887

Title: Increased core temperature following ablation of neurokinin 3 receptor-expressing neurons in the mouse median preoptic nucleus and adjacent preoptic area (MnPO/POA)

Authors: S. J. KRAJEWSKI-HALL, 85724[1], E. M. BLACKMORE[1], J. R. MCMINN[1], N. T. MCMULLEN[2], *N. E. RANCE[1,2,3]
[1]Pathology, [2]Mol. and Cell. Med., Univ. of Arizona Col. of Med., Tucson, AZ; [3]Evelyn F. McKnight Brain Inst., Tucson, AZ

Abstract: We have previously proposed that KNDy neurons play a role in the generation of hot flushes via neurokinin 3 receptor (NK_3R) signaling in the preoptic hypothalamus. This hypothesis is strongly supported by recent clinical studies showing that the number and severity of hot flushes is reduced by treatment with NK_3R antagonists. To determine if preoptic NK_3R neurons modulate thermoregulation in the mouse, we selectively ablated them using injections of saporin conjugated to a NK_3R agonist (NK_3-SAP). NK_3-SAP was stereotaxically injected into Tacr3-EGFP mice to target the MnPO/POA. Controls received injections of BLANK-SAP. The mice were ovariectomized (OVX) and a telemetry probe was implanted i.p. to measure core temperature (T_{CORE}) and activity. Skin temperature (T_{SKIN}) was monitored using a temperature data-logger attached to the surface of the tail. In experiment 1, circadian temperature rhythms were monitored over a 3 day period in mice housed in their home cages (12 light:12 dark). In experiment 2, mice were exposed to three temperatures, 18, 28 and 35ºC, in an environmental chamber. Mice were then implanted s.c. with estradiol (E_2) capsules and the experiments repeated. We verified by immunohistochemistry and quantitative microscopy that approximately 80% of the EGFP-NK_3R neurons in the MnPO were ablated using NK_3-SAP. Ablation of NK_3R neurons significantly elevated T_{CORE} during the light phase in both OVX and OVX + E_2 mice (OVX: BLANK-SAP, 36.7 $\pm$ 0.1 vs NK_3-SAP 37.4 $\pm$ 0.1; OVX+E_2: Blank-SAP 36.1 $\pm$ 0.1 vs NK_3-SAP 36.8 $\pm$ 0.1). NK_3-SAP injections had no significant effect on T_{CORE} during the dark phase. Ablation of NK_3R neurons also increased T_{CORE} during the light phase in mice exposed to 18ºC and 28ºC. All mice exhibited hyperthermia at 35ºC. In contrast, ablation of NK_3R neurons in the MnPO/POA had no effect on T_{SKIN} or activity regardless of experimental treatment. These data suggest that NK_3R neurons in the MnPO/POA participate in the thermoregulatory axis by

promoting heat loss during the day and provide further insight into the CNS thermoregulatory pathways that may be activated during the generation of hot flushes.

Disclosures: S.J. Krajewski-Hall: None. **E.M. Blackmore:** None. **J.R. McMinn:** None. **N.T. McMullen:** None. **N.E. Rance:** None.

Poster

414. Thermoregulation and Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 414.03/PP20

Topic: F.07. Autonomic Regulation

Support: CAPES

CNPq

FAPEMIG

Title: The TRPV1 channel modulates body temperature and cardiovascular responses during physical exercise in rats

Authors: A. C. KUNSTETTER[1], *A. A. ROMANOVSKY[2], W. C. DAMASCENO[1], D. D. SOARES[1], W. PIRES[1], S. P. WANNER[1]
[1]Physical Educ., Federal Univ. of Minas Gerais, Belo Horizonte, Brazil; [2]St Joseph's Hosp. and Med. Ctr., Phoenix, AZ

Abstract: Introduction: The transient receptor potential channel - vanilloid subtype 1 (TRPV1) is a polymodal channel activated by noxious heat, capsaicin and protons. The TRPV1 channel is involved in body temperature regulation in resting rats and in rats exposed to heat. Moreover, this channel mediates metaboreflex and baroreflex responses in rats under general anesthesia. Therefore, in this study, we investigated whether the TRPV1 channel modulates thermoregulatory and cardiovascular responses and performance in rats subjected to a physical exercise.
Methods: The study was approved by a local ethics committee (protocol 31/2014). Male Wistar rats aged between 16-19 weeks were implanted with an abdominal temperature sensor and with a catheter into the ascending aorta artery to measure their core body temperature and blood pressure, respectively. The rats received an intra-arterial injection of a TRPV1 antagonist, N-(4-[6-(4-trifluoromethyl-phenyl)-pyrimidin-4-yloxy]-benzothiazol-2-yl)-acetamide I (AMG 517), at a dose of 60 μg/kg or vehicle (20% ethanol and 80% saline) and then were subjected to a fatiguing, incremental-speed treadmill running at 24°C. During exercise, the rats had their pulsatile arterial pressure, abdominal and tail skin temperatures measured. Mean, systolic and diastolic arterial pressure and heart rate were obtained from the pulsatile arterial pressure

recordings. In addition, the maximal speed attained by the running rats was determined. Results: The injection of AMG 517 increased the abdominal (core) temperature threshold for cutaneous heat loss (38.79 ± 0.30°C vs. 37.66 ± 0.20°C; $p < 0.05$), thus exaggerating the exercise-induced increase in abdominal temperature (39.26 ± 0.43°C vs.38.37 ± 0.38°C; at fatigue; $p < 0.001$) relative to the injection of vehicle. Moreover, the blockade of TRPV1 channels induced a greater increase in mean (130 ± 4 mmHg vs. 120 ± 3 mmHg; at the 30th min; $p < 0.05$) systolic and diastolic arterial pressures. The heart rate increased in both trials, with exaggerated increases being observed during exercise performed after treatment with AMG517. The TRPV1 antagonist did not change the maximal speed attained by the rats (24.2 ± 1.0m/min AMG 517 vs. 22.9 ± 0.7 m/min vehicle; $p = 0.863$).
Conclusions: These data indicate that physiological TRPV1 activation increases cutaneous heat loss, thereby avoiding exaggerated increases in core temperature during exercise. Moreover, TRPV1 activation modulates the exercise-induced increase in arterial blood pressure and heart rate.

Disclosures: A.C. Kunstetter: None. **A.A. Romanovsky:** None. **W.C. Damasceno:** None. **D.D. Soares:** None. **W. Pires:** None. **S.P. Wanner:** None.

Poster

414. Thermoregulation and Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 414.04/PP21

Topic: F.07. Autonomic Regulation

Support: Collins Medical Trust

NIH NS091066

NIH OD010996

Title: A novel neuronal circuit for thermoregulatory inversion

Authors: *D. TUPONE[1,2], G. CANO[3], S. F. MORRISON[4]
[1]Neurosurg., Oregon Hlth. and Sci. Univ., Portland, OR; [2]Dibinem, UNIBO, Bologna, Italy; [3]Univ. of Pittsburgh Dept. of Neurosci., Pittsburgh, PA; [4]Neurolog. Surgery, Oregon Hlth. and Sci. Univ. Dept. of Neurolog. Surgery, Portland, OR

Abstract: We recently described Thermoregulatory Inversion (TI) (DOI: 10.1152/ajpregu.00022.2017), a novel thermoregulatory paradigm, which may be relevant for the induction of hibernation/torpor hypothermia and therapeutic hypothermia to improve outcomes in ischemic pathologies. In contrast to normal thermoregulation, during TI, skin cooling leads to the inhibition of brown adipose tissue (BAT) thermogenesis, whereas skin warming activates

thermogenesis. Here we demonstrate the existence of previously unrecognized, direct neuronal pathways between the dorsal lateral (dl) and the external lateral (el) subdivisions of the parabrachial nucleus (PBN) and the dorsomedial hypothalamus (DMH) that mediate, respectively, the skin cooling-evoked inhibition and the skin warming-evoked activation of BAT thermogenesis during TI. A pre-DMH brain transection induced TI in anesthetized rats that had also received an injection of the retrograde tracer CTb in the DMH. Warm exposure (90 min) of pre-DMH transected rats increased BAT thermogenesis and increased c-fos in DMH and in elPBN neurons projecting to DMH. In anesthetized rats after a pre-DMH transection, inhibition of neurons in either the DMH or the PBN blocked the warm-evoked activation of BAT thermogenesis characteristic of TI. These results are consistent with elPBN neurons relaying warm thermal information to the DMH, resulting in activation of DMH neurons and BAT sympathetic premotor neurons in the rostral raphe pallidus (rRPa) and eliciting a warm-evoked activation of BAT during TI. A population of dynorphinergic neurons in dlPBN projects to DMH, and injection of the k-receptor antagonist (NOR-BNI) in the DMH of pre-DMH transected rats prevents the cold-evoked BAT inhibition during TI. Also, a dynorphin injection in the DMH of intact rats blocks the cold-evoked activation of BAT. This demonstrates the existence of a novel dynorphinergic projection from dlPBN to DMH that may be involved in the inhibition of BAT thermogenesis during TI. We conclude that a novel, short- loop thermoregulatory pathway: skin thermoreceptors → PBN → DMH → rRPa mediates the inverted control of BAT thermogenesis during TI, and that a dynorphinergic input to DMH plays a role in the cold-evoked inhibition of BAT thermogenesis during TI.

Disclosures: D. Tupone: None. **G. Cano:** None. **S.F. Morrison:** None.

Poster

414. Thermoregulation and Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 414.05/PP22

Topic: F.07. Autonomic Regulation

Title: Protection of adiposity during dieting: A role for (lack of)leptin in the dorsomedial hypothalamus

Authors: *R. A. ADAN[1], R. PANDIT[1], A. OMRANI[1], S. E. LA FLEUR[2]
[1]UMCU, Utrecht, Netherlands; [2]Endocrinol. and Metabolism, Academic Med. Center, Univ. of Amsterdam, Amsterdam Zuidoost, Netherlands

Abstract: Weight loss in obesity is hampered by counter regulatory mechanisms such as reduced thermogenesis that can be reversed by injecting leptin peripherally. We discovered that restoring leptin signaling in the dorsomedial hypothalamic nucleus (DMH) is sufficient to normalize the reduced thermogenesis caused by withdrawal from an obesogenic diet. In support of this,

inhibition of leptin signaling in the DMH reduces thermogenesis and promotes adiposity independent of food intake. Leptin's effect on thermogenesis involves DMH neuronal projections to the lateral/dorsolateral periaqueductal grey area. When these neurons are depolarized by leptin or chemogenetically activated by CNO core body temperature and brown adipose tissue thermogenesis increases. These studies collectively demonstrate that a state of relative leptin deficiency during dieting reduces leptin signaling in the DMH resulting in reduced thermogenesis and provides a mechanistic explanation for preservation of adiposity despite lowered caloric intake during dieting.

Disclosures: R.A. Adan: None. **R. Pandit:** None. **A. Omrani:** None. **S.E. La Fleur:** None.

Poster

414. Thermoregulation and Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 414.06/PP23

Topic: F.07. Autonomic Regulation

Support: 1ZIADK07505706

Title: Bombesin-like receptor 3 neurons regulate body temperature, heart rate and food intake

Authors: *R. A. PINOL[1], S. H. ZAHLER[2,1], C. LI[3], B. TAN[3], A. SAHA[3], C. XIAO[3], O. GAVRILOVA[3], A. V. KRAVITZ[4], M. J. KRASHES[5], M. REITMAN[6]
[1]NIH, Natl. Inst. of Diabetes and Digestive, Bethesda, MD; [2]UCSF, San Francisco, CA; [3]NIH, Natl. Inst. of Diabetes and Digestive and Kidney Dis., Bethesda, MD; [4]NIDDK, Natl. Inst. of Hlth., Bethesda, MD; [5]NIH, Bethesda, MD; [6]Diabetes, Endocrinology, and Obesity Br., NIDDK, NIH, Bethesda, MD

Abstract: Bombesin-like receptor 3 (Brs3) is an orphan G protein-coupled receptor expressed in several brain regions, including the dorsomedial hypothalamus (DMH) and paraventricular nucleus of the hypothalamus (PVH). Deletion of Brs3 in mice causes obesity, with reduced energy expenditure and increased food intake, as well as reduced resting body temperature and heart rate in the light phase. Brs3 agonists activate brown adipose tissue (BAT), suppress food intake and cause weight loss. The aim of this project is to identify populations of and circuitry employed by Brs3 neurons that contribute to the regulation of energy expenditure, body temperature, heart rate and food intake. The DMH and PVH have established roles in the regulation of energy expenditure, body temperature, hear rate and in food intake. Nucleus-targeted MK-5046 injections in the DMH, but not in the PVH, raised BAT temperature. Fos activation in DMH^{Brs3} neurons was increased in acutely cold-exposed mice, compared to acutely warm-exposed, but did not change in PVH^{Brs3} neurons. To assess the effect of acute activation of Brs3 populations in freely moving mice we developed Brs3-2A-Cre mice and expressed the

excitatory DREADD hM3D(q). Chemogenetic activation of DMH^{Brs3} neurons increased energy expenditure and body temperature, without influencing physical activity or food intake. In contrast, chemogenetic activation of PVH^{Brs3} neurons had no effect on energy expenditure or body temperature, but suppressed food intake. Optogenetic stimulation of DMH^{Brs3} neurons increased heart rate and blood pressure in addition to body temperature. Accordingly, chemogenetic inhibition of DMH^{Brs3} neurons in reduced the body temperature. DMH^{Brs3} neurons project to the preoptic area, paraventricular nucleus of the hypothalamus, periaqueductal grey and brain stem raphe pallidus (RPa), among other regions. Only optogenetic stimulation of axonal projections from DMH^{Brs3} to the RPa contribute to DMH^{Brs3} neuron mediated thermogenesis. Taken together, the results suggest that the food intake suppression and thermogenic effects of Brs3 activation are mediated by different subsets of Brs3-expressing neurons. Specifically, DMH^{Brs3} neurons can increase energy expenditure, body temperature and heart rate, and PVH^{Brs3} neurons can suppress food intake

Disclosures: R.A. Pinol: None. **S.H. Zahler:** None. **C. Li:** None. **B. Tan:** None. **A. Saha:** None. **C. Xiao:** None. **O. Gavrilova:** None. **A.V. Kravitz:** None. **M.J. Krashes:** None. **M. Reitman:** None.

Poster

414. Thermoregulation and Other

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 414.07/PP24

Topic: F.03. Neuroendocrine Processes

Support: DK025061

Title: Erythropoietin in the brain reduces high fat-diet-induced microglial activation and regulates glucose metabolism

Authors: *S. DEY[1], J. CABAN[1], Z. CUI[2], M. GASSMANN[3], C. T. NOGUCHI[4]
[1]Mol. Med. Br., [2]Mouse Metabolism Core, NIDDK, Bethesda, MD; [3]Inst. of Veterinary(Physiology Vetsuisse Fac., Univ. of Zurich, Zurich, Switzerland; [4]Mol. Med. Branch, NIDDK, Natl. Inst. of Diabetes and Digestive and Kidney Dis., Bethesda, MD

Abstract: High fat-diet (HFD) feeding adversely affects hypothalamic regulation of energy homeostasis. Saturated fatty acids present in HFD can enhance local inflammatory cytokine production and lead to disruption of glucose homeostasis. We hypothesized that Erythropoietin (Epo) signaling in brain could regulate HFD-induced inflammation and thereby maintain glucose and metabolic homeostasis. To study the effect of Epo in the brain, we examined a mouse model of chronic over-expression of human *EPO* transgene in brain (*Tg21*). On normal diet, weights for *Tg21* and wild-type (WT) mice were similar, but Tg21 mice showed significantly improved

glucose tolerance. Insulin levels were not markedly different during Glucose Tolerance Test (GTT) at 0 and 30 minutes for Tg21 and WT mice, but insulin level was significantly lower in Tg21 mice at 120 minutes after glucose injection. When placed on HFD-feeding for 3 weeks, *Tg21* mice gained less fat mass and body weight, and had lower fasting blood glucose compared to WT mice. Moreover, *Tg21* mice on HFD showed improved glucose tolerance compared to WT mice in GTT assay. Insulin levels at baseline were similar in *Tg21* and WT mice, increased transiently at 30 min after glucose injection with *Tg21* mice reaching a level twice that of WT mice, and returning to baseline at 120 minutes. Immunofluorescent staining of the arcuate nucleus region of hypothalamic sections showed lower inflammatory cytokine TNFα and activated microglia marker Iba1 in *Tg21* compared to WT mice. To determine the direct response in brain to elevated EPO, WT mice underwent acute intracerebroventricular administration of Epo (Epo ICV). EPO ICV prevented HFD-induced fat mass and weight gain relative to saline-controls (saline ICV), although no difference in food intake was observed. Epo ICV mice also showed lower fasting blood glucose. Hematocrits were unchanged compared with saline ICV, suggesting no peripheral effects of Epo ICV. Epo ICV mice showed lower hypothalamic gene expression for inflammatory markers *TNFα, IL6, IL1α, SOCS3* and higher expression of anti-inflammatory *IL10*. In summary, our studies suggest an important regulatory role of Epo in the brain in preventing HFD fat gain, glucose homeostasis, not mediated by a primary alteration in insulin production, and in protecting against HFD associated brain inflammation and microglial cell activation in the hypothalamus.

Disclosures: S. Dey: None. **J. Caban:** None. **Z. Cui:** None. **M. Gassmann:** None. **C.T. Noguchi:** None.

Poster

415. Early Life and Intergenerational Effects on Feeding

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 415.01/PP25

Topic: F.10. Food Intake and Energy Balance

Support: UCNI-NRC Pilot Project Grant

MH106330

Title: Perinatal high fat diet leads to DNMT activity deficits in the offspring prefrontal cortex

Authors: *K. R. LLOYD[1], S. E. MCKEE[3], N. M. GRISSOM[4], B. L. SMITH[2], T. M. REYES[2]
[2]Psychiatry and Behavioral Neurosci., [1]Univ. of Cincinnati, Cincinnati, OH; [3]Univ. of Pennsylvania, Philadelphia, PA; [4]Dept. of Psychology, Univ. of Minnesota, Minneapolis, MN

Abstract: Excessive gestational weight gain and maternal obesity increase the risk of neurological disorders, including schizophrenia, autism, and attention deficit/hyperactivity disorder (ADHD) in offspring. These disorders of cognition and attention implicate the prefrontal cortex (PFC), a region important for action selection, executive function, and inhibitory control. Previously, we have shown that offspring of a maternal high-fat diet (HFD) have whole-genome DNA hypomethylation, promoter-specific hypomethylation and dysregulated gene expression in the PFC, suggesting that DNA methylation deficits may contribute to the neurodevelopmental programming deficits induced by gestational exposure to a HFD. We were interested in understanding whether HFD offspring had specific deficits in DNA methyltransferase expression, localization or function which may contribute to the risk for neurodevelopmental programming deficits. Using a mouse model of maternal overnutrition, we identified decreases in overall DNMT activity due to maternal HFD in offspring, without changes in RNA gene expression or protein levels. We provide evidence for cytoplasmic sequestration of DNMT1 proteins, as cytoplasmic sequestration of DNMT1 has been observed in the PFC in schizophrenia and dementia. However, immunoprecipitation experiments revealed no difference in the binding of DNMT1 to kinases which regulate DNMT1 nuclear import and catalytic function. Overall, these findings demonstrate that early life nutrition can permanently alter the function of DNMT1 into adulthood, possibly predisposing to neurological disorders.

Disclosures: K.R. Lloyd: None. **S.E. McKee:** None. **N.M. Grissom:** None. **B.L. Smith:** None. **T.M. Reyes:** None.

Poster

415. Early Life and Intergenerational Effects on Feeding

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 415.02/PP26

Topic: F.10. Food Intake and Energy Balance

Support: R01AA024798-01

Title: Embryonic exposure to ethanol in zebrafish: Effects on development and migration of hypothalamic orexin neurons

Authors: *A. EVANS, E. RAMIREZ, V. HALKINA, V. KEWALDAR, G.-Q. CHANG, O. KARATAYEV, S. F. LEIBOWITZ
Rockefeller Univ., New York, NY

Abstract: Embryonic ethanol is known to increase alcohol drinking in humans and animals and have long-term effects on neurochemical systems in rodent brain. In our recent paper, we demonstrated a similar phenomenon in adult zebrafish (ZF), where voluntary consumption of an ethanol-gelatin meal significantly stimulates the orexigenic neuropeptide, orexin/hypocretin

(ox/hcrt), in the anterior hypothalamus (AH) and also behaviors induced by this peptide. Additionally, embryonic exposure to ethanol in water compared to control stimulates neurogenesis in the AH, the proliferation and expression of neurons expressing ox/hcrt, and voluntary consumption of 10% ethanol-gelatin in adult ZF. In this study, we further characterized this model of 10% ethanol-gelatin feeding and found that the daily number of bites taken by the ZF of an ethanol-gelatin meal in 5 minutes was the most reliable, day-to-day measure and strongly positively correlated with blood alcohol levels ($r=+0.73$, $p<0.05$). Daily consumption of an ethanol-gelatin meal for 4 weeks compared to plain gelatin significantly increased novelty-induced locomotor behavior and aggression in the adolescent offspring and neurogenesis and density of neurons expressing ox/hcrt in the AH, as revealed by immunofluorescence histochemistry and *in situ* hybridization. To further understand how early ethanol exposure affects the development of ox/hcrt neurons that promote ethanol intake and related behaviors, we utilized live imaging techniques to characterize the proliferation and migration of these neurons in the hcrt:EGFP transgenic fish. Using Imaris software, examination of larvae exposed to 0.5% ethanol in water from 22-24hpf and imaged from 24 to 28hpf revealed a novel finding, that ethanol compared to water control strongly hinders the movement of ox/hcrt cells within the developing hypothalamus. In response to ethanol, the ox/hcrt cells traveled a shorter net distance within the 4 hour imaging period, as indicated by a decrease in displacement length ($p<0.03$). They also migrated in a disorganized manner, as indicated by the reduced straightness of their individual migratory paths ($p<0.01$). Further, preliminary data suggest that the horizontal movement of ethanol-exposed cells differs from controls, with the latter moving away from the midline while the former migrate towards it. This demonstrates that embryonic exposure to ethanol in ZF markedly affects the brain and behavior of larval and adult offspring, providing new evidence for ethanol-induced changes in the migratory path of ox/hcrt cells in developing hypothalamus that may affect their ultimate location and contribute to the behavioral disturbances.

Disclosures: **A. Evans:** None. **E. Ramirez:** None. **V. Halkina:** None. **V. Kewaldar:** None. **G. Chang:** None. **O. Karatayev:** None. **S.F. Leibowitz:** None.

Poster

415. Early Life and Intergenerational Effects on Feeding

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 415.03/PP27

Topic: F.10. Food Intake and Energy Balance

Support: PAPIIT IN212516 CONACYT- Fronteras de la Ciencia 398

Title: Effects of maternal over-nutrition on offspring´s cognitive processes and behavioral patterns

Authors: *Y. BARB[1], E. NAVARRETE[1], R. MONTÚFAR-CHAVEZNAVA[2], A. SALAZAR[1], P. OSTROSKY-SHEJET[1], I. CALDELAS[1]
[1]Inst. De Investigaciones Biomédicas, UNAM, Ciudad de México, Mexico; [2]Facultad de Ingeniería, Univ. Nacional Autónoma de México, Ciudad de México, Mexico

Abstract: Chronic malnutrition during pregnancy has been associated to structural, physiological and metabolic changes, as well as behavioral impairments in the offspring. It is critical to elucidate whether chronic over-nutrition induced by the intake of a high lipids and carbohydrates diet during pregnancy, affect the cognitive processes and behavioral patterns of the offspring. In order to address this issue, a Food Preference Test (FPT) and a Novel Object Recognition Test (NOR), were performed on six months old European rabbits obtained from females fed with a standard diet (CON) or with a high lipids and carbohydrates diet during gestation (HLCD). In order to asses if the HLCD rabbits had a predisposition for unbalanced food, FPT was performed with a total of 47 rabbits. In order to evaluate the effect of the nutritional status during embryonic development on short-term declarative memory of offspring, the NOR test was performed with a total of 40 rabbits.
In the FPT, animals from the HLCD group ingested significantly more food than the rabbits from the CON group. HLCD males required more time to choose one of the diets. However, the sequence of behaviors associated with food consumption was similar between both groups, with the exception of grooming, that exhibited a significant increase in the frequency in the HLCD animals. The grooming and urination frequency was greater in males than in female rabbits. In the NOR test, the HLCD rabbits exhibited a significant increase on the latency of exploration of the novel object.
Preliminary results from the FPT indicate that the subjects obtained from over-nourished females exhibit hyperphagia, however, they did not show a preference for high fat/carbohydrates diets, as suggested in studies in other species. The significant increase in the grooming behavior presented by the rabbits of the HLCD group, in the FPT test, may be considered as an indicator of the presence of stereotyped and/or compulsive behaviors. In addition, the HLCD rabbits require more time to exhibit a proper response to novel objects. Further studies are needed to determine with more accuracy the effects of maternal nutrition on cognitive processes.

Disclosures: Y. Barb: None. **E. Navarrete:** None. **R. Montúfar-Chaveznava:** None. **A. Salazar:** None. **P. Ostrosky-Shejet:** None. **I. Caldelas:** None.

Poster

415. Early Life and Intergenerational Effects on Feeding

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 415.04/PP28

Topic: F.10. Food Intake and Energy Balance

Support: NIh Grant R01 DA025674-04A1

Title: Female adolescent opioid exposure confers vulnerability to metabolic dyshomeostasis in F1 offspring in a diet-specific context

Authors: *A. TOORIE[1], F. M. VASSOLER[2], A. KUBEREK[3], C. WYSE[3], E. M. BYRNES[4]
[1]Biomed. sciences, Tufts Univ., North Grafton, MA; [2]Dept. of Biomed. Sci., Tufts Univ. Cummings Sch. of Vet. Med., North Grafton, MA; [4]Biomed. Sci., [3]Tufts Univ. Cummings school of Vet. Med., North Grafton, MA

Abstract: In the US, opioid abuse is a current major public health issue. Opioids regulate reward- and motivation- related behaviors and chronic opioid use results in dysfunctions in the brain's reward system. Opioid misuse/abuse amongst adolescents has increased significantly; yet, the long-term consequences of this experience has yet to be realized. Data from animal models, however, reveal that adolescently opioid-exposed females transmit to F1 offspring transgenerational neurobehavioral and endocrine changes at both baseline and in the context of various stimuli. For example, the offspring of females that were exposed to morphine during adolescence (Mor-F1) display significant differences in hypothalamus-pituitary-adrenal axis function, and increased hypothalamic ARC proopiomelanocortin (*POMC*) gene expression in a sex-specific manner. In the brain, POMC is the precursor to several neuropeptides involved in homeostatic and adaptive processes. Two bioactive derivatives of POMC, β-EP and α-MSH, are central regulators of energy homeostasis and metabolism. In the present study, Mor-F1 and Sal-F1 (maternal history of adolescent saline exposure; control) animals were maintained under different dietary conditions (control, high fat diet, or high sucrose diet) to assess neuroendocrine and metabolic changes. Specifically, we tested the hypothesis that Mor-F1 offspring are vulnerable to the development of metabolic dyshomeostasis when challenged with excessive fat or sugar. In the current study, Mor-F1 offspring displayed increased ARC POMC levels alongside alterations in β-EP and α-MSH levels within the ventral tegmental area, in a diet-specific manner. F1 animals also displayed differences in fasting glucose levels, alongside changes in the glucoregulatory hormones, insulin and corticosterone. In addition, Mor-F1 animals sex-specifically displayed changes in sexual maturity. Collectively, results demonstrate sex-dependent alterations to neuroendocrine systems that facilitate development and metabolism as a function of maternal opioid use during adolescence. Moreover, results suggest that female adolescent opioid exposure increases the risk of diet-induced metabolic derangement (i.e., weight gain, fasting hyperglycemia) in F1 progeny. Current studies are examining the underlying mechanisms responsible for bodyweight gain and glucose dyshomeostasis in F1 offspring.

Disclosures: A. Toorie: None. **F.M. Vassoler:** None. **A. Kuberek:** None. **C. Wyse:** None. **E.M. Byrnes:** None.

Poster

415. Early Life and Intergenerational Effects on Feeding

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 415.05/PP29

Topic: F.10. Food Intake and Energy Balance

Support: FAPESP

CAPES

CNPq

Title: Neonatal nutritional programming and hypothalamic glial cell morphology: the role of leptin in the modulation of TCPTP and conexins

Authors: *L. K. DEBARBA[1], P. B. MARANGON[2], F. M. V. VECHIATO[2], H. V. P. SILVA[2], J. C. VENÂNCIO[2], B. C. BORGES[2], G. ALMEIDA-PEREIRA[2], M. C. JAMUR[3], J. ANTUNES-RODRIGUES[2], L. L. K. ELIAS[2]
[2]Physiol., [3]Cell and Mol. Biol. and Pathogenic Bioagents, [1]Ribeirão Preto Med. Sch. / USP, Ribeirao Preto, Brazil

Abstract: Nutritional changes in the neonatal period induce energy balance impairment in the juvenile and adult life. Astrocytes provide energy substrate to the neurons through conexin 43 (CX43). CX30 exerts a role in the maintenance of astrocyte morphology. The T-cell protein tyrosine phosphatase (TCPTP) is a counter-regulatory molecule of leptin signaling which also modulates CX43 expression. We used the neonatal nutritional programming model by altering the litter size during lactation in rats: 3 puppies - Small Litter (SL); 10 puppies - Normal Litter (NL) and 16 puppies per dam - Large Litter (LL). The SL animals showed higher body weight at weaning and thereafter until the 60th post-natal day (PN). LL animals reduced body weight gain at weaning, however they reached the same body weight gain of NL animals at PN35, and this effect was associated with hyperphagic behavior and increased *Il1b* mRNA expression in the arcuate nucleus (ARC). In the SL (PN21) and in the SL and LL (PN60) there were an increase in: a) leptin and insulin plasma concentrations; b) ARC *Ptpn2* (TCPTP encoding gene) mRNA expression; c) hypothalamic TCPTP protein content; d) ARC GFAP and IBA1 immunoexpression and e) TCPTP overlap with GFAP in the ARC. In the SL at PN21 an increase in ARC CX43 immunoexpression was observed while at PN60 there was a reduction of CX30 immunoexpression and an increase of *Tnf* mRNA expression in the ARC in both groups, SL and LL. At PN21 SL showed an increase in the soma and, also, in the process extension of the microglia and astrocytes in the ARC. Similar result was observed in ARC astrocytes of both SL and LL groups at PN60. Using primary culture of rat hypothalamic astrocytes, we observed that the leptin [5000ng / mL] increased the *Ptpn2* mRNA expression in the, as well as, an increase of

TCPTP and CX43 immunoexpression and a reduction of CX30 immunoexpression. These effects of leptin were associated with a marked increase of astrocyte soma. Moreover, the silencing of *Ptpn2* (siRNA *Ptpn2*) was able to reverse all these results promoted by leptin. The data demonstrate that neonatal nutritional changes induce long-lasting alterations in the energy balance in the juvenile and adult life. These changes are associated with morphological changes of glial cells and an increase of proinflammatory cytokines in the ARC. Leptin appears to play a role in the alterations of astrocyte morphology regulating the expression of TCPTP, which in turn modulates the expression of gap junction protein CX30.

Disclosures: L.K. Debarba: None. **P.B. Marangon:** None. **F.M.V. Vechiato:** None. **H.V.P. Silva:** None. **J.C. Venâncio:** None. **B.C. Borges:** None. **G. Almeida-Pereira:** None. **M.C. Jamur:** None. **J. Antunes-Rodrigues:** None. **L.L.K. Elias:** None.

Poster

415. Early Life and Intergenerational Effects on Feeding

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 415.06/QQ1

Topic: F.10. Food Intake and Energy Balance

Support: FAPESP Grant 2016/13136-6

CAPES

Title: Effects of perinatal programming by caloric restriction on the cocaine and amphetamine regulated transcript (CART) neurons in the lateral hypothalamic area of weaned rats

Authors: *C. D. MACHADO[1], M. G. MARTINS[1], I. Z. GUIATI[1], J. C. BITTENCOURT[2], J. A. C. HORTA, Jr[1]
[1]Anat., Inst. of Biosci. of Botucatu, Botucatu, Brazil; [2]Anat., Inst. of Biomed. Sci., Sao Paulo, Brazil

Abstract: Perinatal programming is related to adaptations performed in response to insults during key points of development, leading to effects in organs' structure and function. Maternal nutritional status during pregnancy and lactation is considered an important programming inductor, since nutrient deficiency causes significant changes in body weight, energy balance, food intake and neuropeptides expression. The cocaine and amphetamine regulated transcript (CART) is a neuropeptide widely expressed in the hypothalamus. In the lateral hypothalamic area (LHA), it is involved in the energy balance, playing an anorexigenic role. Our aim was to analyze the impact of maternal perinatal undernutrition on the number, distribution and ultrastructure of CART-neurons in the LHA of weaned rats. Two groups of pregnant *Wistar* rats (Ethical Committee Approval nº722) were studied: control group (CG), *ad libitum* standard diet,

and caloric restricted group (RG), 50% restriction in diet compared to the control group during pregnancy and lactation. Male pups in postnatal day 21 (N=8/group) were analyzed. All animals were transcardially perfused with fixative and their brains processed for CART-neurons analysis in the LHA, according to immunohistochemistry protocols (N=5) and transmission electron microscopy (N=3). Body weight, body length, brain and adipose tissue (visceral and retroperitoneal) weights and glycemia were also evaluated. The mapping and estimation of CART-neurons number in the LHA were performed using 3D reconstruction and stereology. Quantitative data were compared and statistically analyzed ($p < 0.05$). RG animals showed a decrease in body weight and length. Glycemia, brain, visceral and retroperitoneal adipose tissue also reduced in RG animals. There were fewer CART-neurons in LHA in RG, however there were no differences in LHA total area and volume between groups. CART-neurons distribution in LHA was similar both in CG and RG animals. Positive-labeled neurons were identified mainly in mamilar and tuberal LHA regions, and in dorsolateral, perifornicial and ventrolateral LHA sub-regions. No relevant differences between groups were found in CART-neurons ultrastructure. Nuclear membrane invagination and labeled Golgi apparatus with dense granules, typically found in peptidergic neurons, were observed. Granular endoplasmic reticulum cisternae and multivesicular bodies were also labeled. Our results indicated that maternal perinatal undernutrition induced body structural changes and decreased CART-neurons in the LHA of weaned rats. Nevertheless, LHA total area and volume, CART-neurons ultrastructure and distribution were not modified.

Disclosures: **C.D. Machado:** None. **M.G. Martins:** None. **I.Z. Guiati:** None. **J.C. Bittencourt:** None. **J.A.C. Horta:** None.

Poster

415. Early Life and Intergenerational Effects on Feeding

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 415.07/QQ2

Topic: F.10. Food Intake and Energy Balance

Support: CONACyT grant FOSSIS 233918

Title: Effect of prenatal stress on adrenal and thyroid axes of adult male rats subjected to light-phase feeding during prepuberty

Authors: *C. GARCIA-LUNA, P. SOBERANES-CHAVEZ, P. DE GORTARI
Inst. Nacional De Psiquiatria, Mexico city, Mexico

Abstract: Obesity development involves interaction between factors including feeding patterns and stress. Neuronal circuits responsible for energy homeostasis regulation and stress response mature during gestation and early postnatal stage, thus fluctuations in serum hormones levels

implicated in those functions such as leptin, insulin, thyroid hormones and corticosterone (GC) could alter the programmed regulation of neuroendocrine axes and limbic neural nets leading to hyperphagia and overweight that might last up to adulthood. Prenatal stress (PS) induces high GC serum content that affects development of the hypothalamic-pituitary-adrenal (HPA) and hypothalamic-pituitary-thyroid (HPT) axes that could lead to an aberrant HPA axis response to stress and to an impaired regulation of basal energy expenditure by HPT axis later in life. Prepuberal un-stressed animals subjected to time-restricted feeding schedules (tRF) during rest phase present hyperphagia and overweight up to adulthood due to a changed energy homeostasis regulation that was associated to hyperactivity of HPA axis and inhibition of HPT axis function. PS induces neuroendocrinology impairments by altering neural development, thus we analyze if PS is able to exacerbate prepuberal tRF-induced changes in HPT and HPA axis function of adult rats. Pregnant Wistar rats were subjected (PS-C) or not (C) to chronic mild stress on the last week of gestation, their male offspring divided and subjected to light-phase feeding (PS-LPF) or dark-phase feeding (PS-DPF) from 21-35 postnatal (PN) days; after that, they had *ad libitum* access to food until day 70PN. Body weight and food intake were measured weekly. Rats were sacrificed at week 10, trunk blood collected to evaluate GC, thyroid hormones (TH), leptin and insulin content; brain excised and kept at -70°C to analyze PVN TRH and CRF RNAm expression by ISH. Body weight and food intake did not differ between PS groups although they were higher than C animals. TH content did not show differences between PS groups or vs C suggesting an inhibition of HPT axis that could favor overweight; GC levels were higher than C but did not differ among PS rats showing HPA axis hyperactivation by PS, leptin and insulin content was higher vs C relating them to the overweight and suggesting leptin and insulin resistance since PS rats showed hyperphagia and those hormones are anorexigenic agents. PVN TRH and CRF mRNA expression did not differ between PS groups or vs C. In conclusion, PS did not exacerbate changes caused by tRF; both conditions caused hyperphagia, overweight, HPA axis hyperactivity, non-responsive HPT axis to positive energy balance, and a possible insulin and leptin resistance.

Disclosures: C. Garcia-Luna: None. **P. Soberanes-Chavez:** None. **P. de Gortari:** None.

Poster

415. Early Life and Intergenerational Effects on Feeding

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 415.08/QQ3

Topic: F.10. Food Intake and Energy Balance

Support: FACEPE Grant APQ-0164-4.05/15

Title: Energy balance in overfeed rats

Authors: A. DA SILVA[1], G. F. BRAZ[2], S. A. SILVA[2], C. M. FREITAS[2], *C. LAGRANHA[2]
[1]UFPE, Recife, Brazil; [2]CAV-UFPE, Vitoria DE Santo Antao, Brazil

Abstract: Obesity is basically induced by an imbalance between energy intake and energy expenditure. Overweight has becoming increasingly prevalent in children's and teenagers. According to WHO, obesity is an epidemic disease and the subsequent morbidity and mortality are the major health challenge worldwide. Early postnatal overnutrition in rats induce a rapid grow, weight gain, fat deposition, hyperphagia during lactation and hypertriglyceridemia in adulthood. Several antidepressants such as selective serotonin reuptake inhibitors (i.e fluoxetine also known as Prozac) are now prescribed not only for depression, but also to obesity treatment. With this our aim was to evaluate the effect of fluoxetine treatment during 21 days (from 39 to 59 days of age) in the control of energy balance. To induce neonatal overnutrition, the primary litter size was then reduced on the third day of life to only three pups per litter (SL; n =8). In normal litters, the size was adjusted to 9 neonates per mother (NL; n =9). The fluoxetine treatment started with 39 days of age, using 10mg/Kg B.W. (i.p injection) in male Wistar rats. At 60 days of age was evaluated blood parameters, 24 hours food intake, 12 hours in dark cycle food intake, 12 hours in light cycle food intake, food intake after 3 hours of food-deprived, body weight, white and brown adipose tissue weight, POMC and NPY mRNA. Our result showed that neonatal overnutrition induces body weight increase, white adipose tissue weight, increases triglycerides, but no differences were observed in food intake in basal situation, but after 3 hours of food-deprivation, SL group eat 40% more than NL. Additionally, neonatal overnutrition has higher NPY expression than NL. After fluoxetine treatment, we observed decreases in body weight, white adipose tissue weight, triglycerides, food intake after food-deprivation and NPY expression combined with increase in POMC expression. Our results suggest that fluoxetine treatment modulates energy balance in obese rats, mainly by control food intake after restriction and fat deposition.

Disclosures: A. da Silva: None. **G.F. Braz:** None. **S.A. Silva:** None. **C.M. Freitas:** None. **C. Lagranha:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.01/QQ4

Topic: F.10. Food Intake and Energy Balance

Support: NS38809

NS43330

DK68098

Title: Estradiol protects proopiomelanocortin neurons against insulin resistance

Authors: *J. QIU[1], M. A. BOSCH[1], U.-V. NAVARRO[1], C. C. NESTOR[1], O. K. RØNNEKLEIV[1,2], M. J. KELLY[1,2]
[1]Oregon Hlth. and Sci. Univ., Portland, OR; [2]Oregon Natl. Primate Res. Ctr., Beaverton, OR

Abstract: The incidence of metabolic syndrome sharply increases in women after menopause. Neurons, similar to fat and muscle cells, can develop hyperinsulinemia-induced IR, which results in severe injury to the nervous system as seen in diabetic neuropathies and stroke. At the center of the regulation of energy homeostasis, and hence the central feedback of insulin, are the anorexigenic proopiomelanocortin (POMC) and orexigenic neuropeptide Y/agouti-related peptide (NPY/AgRP) neurons in the hypothalamic arcuate nucleus. Since 17β-estradiol (E_2) augments the excitability of the anorexigenic POMC neurons, we investigated the electrophysiological effects of insulin in diet-induced obese (DIO) male and female mice, and the neuroprotective effects of E_2 in females. We found that the ability of insulin to activate canonical transient receptor potential 5 (TRPC5) channels and depolarize POMC neurons was significantly reduced in DIO male (by 70%) but not in DIO female mice. However, the insulin response in POMC neurons was attenuated in ovariectomized, DIO females but restored with E_2 replacement. E_2 increased T-type calcium channel *Cav3.1* mRNA expression and currents but downregulated stromal-interaction molecule1 (*Stim1*) mRNA, which rendered POMC neurons more excitable and responsive to insulin-mediated TRPC5 channel activation. Also, in females E_2 prevented the increase in *Socs3* mRNA expression with DIO that was seen in males. Therefore, E_2 protects female POMC neurons from insulin resistance with DIO by enhancing POMC neuronal excitability and the coupling of insulin receptor to TRPC5 channel activation.

Disclosures: J. Qiu: None. **M.A. Bosch:** None. **U. Navarro:** None. **C.C. Nestor:** None. **O.K. Rønnekleiv:** None. **M.J. Kelly:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.02/QQ5

Topic: F.10. Food Intake and Energy Balance

Support: NIH Grant T32

Title: Genetic dissection of leptin regulated neural circuits controlling energy balance and glucose homeostasis

Authors: *C. L. BARTOLOME[1], J. XU[1], C. S. LOW[1], X. YI[1], C.-H. CHIEN[1], P. WANG[1], D. KONG[2]
[1]Neurosci., Tufts Univ., Boston, MA; [2]Tufts Univ. Sch. of Med., Boston, MA

Abstract: Leptin is a hormone released from adipose tissue that works exclusively in the brain to regulate energy balance and glucose homeostasis. Dysfunctions in leptin or leptin signaling induce severe obesity and diabetes. Since the discovery of leptin in 1994, various efforts have been made to identify the first-order leptin-responsive neurons that mediate its anti-obese and anti-diabetic effects. Recent studies have shown that leptin completely rescues streptozotocin-induced type 1 diabetes (T1D) in an insulin- independent manner. Using this model, we postulated that STZ-induced T1D induces altered neuronal activity in the CNS, which can be reversed upon leptin treatment. Surprisingly, we found over 50 sites with increased neuronal activity, and identified a small group of hypothalamic neurons that acutely reversed its neuronal firing upon leptin treatment. Furthermore, we inhibited these hypothalamic neurons using DREADDs in STZ-treated mice and found that acute neuronal inhibition completely attenuated the diabetic hyperphagia and dramatically reduced hyperglycemia. To assess whether the increased neuronal activity was due to a lack of leptin in an STZ mouse model, we utilized the CRISPR/Cas9 system to efficiently knockout leptin receptors in these hypothalamic neurons of adult mice. Leptin receptor deletion induced 81% of the severe obesity phenotype observed in *db/db* mice, suggesting that these hypothalamic neurons are a primary target of leptin to regulate energy balance and glucose homeostasis. These findings provide an innovative framework of brain-based mechanisms controlling energy balance and glucose homeostasis, provide novel information towards the understanding and treatment of obesity and diabetes, and bring about potential brain- related therapeutic strategies to treat severe metabolic disorders.

Disclosures: C.L. Bartolome: None. **J. Xu:** None. **C.S. Low:** None. **X. Yi:** None. **C. Chien:** None. **P. Wang:** None. **D. Kong:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.03/QQ6

Topic: F.10. Food Intake and Energy Balance

Support: NIH DK704734

NIH DA035088

Title: Behavioral, anatomical and molecular investigation of PACAP and leptin interactions in the rat ventromedial hypothalamus

Authors: *M. M. HURLEY, E. M. HESS, B. MAUNZE, C. LAMBERTON, M. FRENKEL, G. CALLAN, N. S. PATEL, D. A. BAKER, S. CHOI
Biomed. Sci., Marquette Univ., Milwaukee, WI

Abstract: Obesity is rising at an alarming rate in the United States of America and it is critical that we address this growing crisis as this disease puts great socioeconomic strain on our society. Leptin, a hormone that is produced peripherally by adipose cells, has been shown to play an important role in regulating mammalian energy states. As obese individuals demonstrate impairments in leptin mediated signaling, it remains critical to continue to interrogate the mechanisms by which leptin regulates normal and pathological feeding states. Leptin has been shown to act centrally in the ventromedial hypothalamic nuclei (VMN). Previous work has demonstrated that leptin microinjected into the rat VMN, strongly suppresses feeding and increases phosphorylation of signaling transducers and activators of transcription 3 (P-STAT3) as well as brain derived neurotrophic factor (BDNF) and suppressor of cytokine signaling 3 (SOCS3) mRNA expression. Our current data demonstrate that the secretin family neuropeptide pituitary adenylate cyclase-activating polypeptide (PACAP), much like leptin, also decreases feeding behavior, body weight gain and increases phosphorylation of STAT3 and BDNF/SOCS3 mRNA expression when injected into the VMN. To assess if both leptin and PACAP are signaling through co-dependent molecular mechanisms in the VMN, we antagonized VMN PACAP receptors with PACAP6-38 followed immediately by a leptin microinjection. We observed that animals pretreated with PACAP6-38 did not display leptin induced hypophagia. This interesting behavioral effect suggests that endogenous PACAP receptor-dependent activity is necessary for leptin-induced suppression of feeding behavior in the VMN. To further validate the relationship between PACAP and leptin signaling in the VMN, we conducted double fluorescent *in situ* hybridization which showed that PAC1 and leptin receptor mRNA transcripts are co-expressed in the same cells within the VMN. Taken together, these data suggest a functional link between PACAP and leptin signaling, and possibly BDNF, in the VMN in the regulation of feeding behavior and metabolism. Future studies will further explore the functional interactions between leptin, PACAP, and BDNF in the regulation of energy homeostasis and feeding behavior.

Disclosures: M.M. Hurley: None. **E.M. Hess:** None. **B. Maunze:** None. **C. Lamberton:** None. **M. Frenkel:** None. **G. Callan:** None. **N.S. Patel:** None. **D.A. Baker:** None. **S. Choi:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.04/QQ7

Topic: F.10. Food Intake and Energy Balance

Support: CIHR Grant MOP12192

Title: Cellular actions of adropin on paraventricular nucleus neurons

Authors: *S. P. LOEWEN, A. V. FERGUSON
Dept. of Biomed. and Mol. Sci., Queen's Univ., Kingston, ON, Canada

Abstract: Adropin is a peptide hormone that has been observed to have metabolic roles in the periphery and a central role to inhibit water intake. We have recently shown that adropin directly influences the excitability neurons in the hypothalamic paraventricular nucleus (PVN). The current study uses electrophysiological techniques to examine the responsiveness of PVN neurons to adropin at different extracellular glucose concentrations and determine the ionic mechanisms involved. In our original experiments at 10 mM glucose (central hyperglycemia), adropin (10 nM) elicited responses in 68% of cells tested (n=57/84). The majority of cells (58%) depolarized (5.2 ± 0.3 mV; n=49) in response to adropin, while the remaining responsive cells (10%) hyperpolarized (−3.4 ± 0.5 mV; n=8). Here, we show that reductions in the extracellular glucose concentration eliminates hyperpolarizing responses. In 3 mM glucose (central normoglycemia), adropin (10 nM) elicited responses in 47% of cells tested (n=7/15), all of which were depolarizations (4.8 ± 0.5 mV). In 1 mM extracellular glucose (central hypoglycemia), 10 nM adropin elicited depolarizations in 67% of cells tested (6.0 ± 0.8 mV; n=8/12), while the remaining 33% of neurons (n=4) did not respond to adropin application. The magnitudes of adropin-mediated depolarizations did not differ at each glucose concentration (one-way ANOVA, p=0.50). Additionally, analysis of action potential shape in neurons that depolarized revealed a broadening of spikes after adropin application, suggesting adropin may affect a potassium conductance during these responses. These findings demonstrate that the glucose environment affects adropin-mediated responses in PVN neurons and reveal the importance of considering glycemic state when studying molecules or pathways pertaining to the central regulation of energy homeostasis.

Disclosures: S.P. Loewen: None. **A.V. Ferguson:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.05/QQ8

Topic: F.10. Food Intake and Energy Balance

Support: Clayton Medical Research Foundation

Title: In search of the cocaine- and amphetamine-regulated transcript (CART) receptor: Identification of CART binding sites in the rodent brain

Authors: *L. A. TAN, M. H. PERRIN, J. M. VAUGHAN, C. M. ARIAS, K. A. LEWIS, J. E. RIVIER, P. E. SAWCHENKO
LNSF-S, Salk Inst. For Biol. Studies, La Jolla, CA

Abstract: Cocaine- and amphetamine-regulated transcript (CART) is an endogenous peptide that has been implicated in the regulation of food intake and reward. CART exists in two forms, CART(55-102) and CART(61-102), with a highly conserved structure governed by three disulfide bridges. CART is abundantly expressed in many areas of the brain associated with the reward circuit and the control of feeding and stress responses, as well as peripherally, in the adrenal and pituitary glands and GI tract. CART satisfies many of the requirements of a neurotransmitter, and injections of CART into the brain reduce food intake and inhibit the effects of psychostimulants. Despite a wealth of literature on the physiological roles of CART peptides, no putative CART receptor has been identified, although binding sites have been reported on PC-12 and AtT-20 cell lines and nucleus accumbens primary neurons. To facilitate identification of CART receptors in rodent brain, we developed a new CART analog, [^{125}I-Tyr^{58},Thr^{62},Nle^{67}]CART(55-102), and demonstrated its high affinity (low nanomolar), specific binding to membranes from porcine anterior pituitary, rodent brain and pancreatic-, neuronal- and adrenal-derived cell lines. Because CART is present at high levels in rat plasma (333-675 pg/ml), we used this tracer to gain clues regarding receptor disposition by identifying potential source(s) and target(s) of circulating peptide. Neither adrenalectomy nor hypophysectomy affected plasma peptide levels, while sleeve gastrectomy did, implicating the GI tract as the major source of circulating CART. Following i.v. injection, however, we failed to resolve time-dependent sequestration of the tracer in the brain or any peripheral tissue, as measured by gamma counting or SPECT imaging. Taking advantage of the resolution afforded by *ex vivo* autoradiography, we were able to demonstrate robust, specific binding in rat and mouse brain that was competable in a dose-related manner by unlabeled CART. Such binding was restricted to secretory structures including the paraventricular and supraoptic nuclei of the hypothalamus, choroid plexus, and anterior pituitary, and did not parallel the widespread central distribution of CART projections, or include such demonstrated sites of peptide action as the nucleus accumbens. These findings provide the first autoradiographic localization of CART binding sites in brain and pituitary. This information, and the novel radioligand that fostered it, should provide leverage in further efforts to identify the CART receptor(s).

Disclosures: L.A. Tan: None. **M.H. Perrin:** None. **J.M. Vaughan:** None. **C.M. Arias:** None. **K.A. Lewis:** None. **J.E. Rivier:** None. **P.E. Sawchenko:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.06/QQ9

Topic: F.10. Food Intake and Energy Balance

Title: Intracellular signaling involved in neurokinin NK1 receptor-mediated emesis in the least shrew

Authors: W. ZHONG[1], S. CHEBOLU[2], *N. A. DARMANI[2]
[1]Basic Med. Sci., [2]Coll Osteo. Med. Pacific, Western Univ. Hlth. Sci., Pomona, CA

Abstract: Previously we have found a major CNS emetic component for the neurokinin 1 receptor (NK1R)-mediated vomiting evoked by the selective NK1R agonist GR73632 (5 mg/kg, i.p.) in the least shrew. The evoked vomiting was suppressed in a dose-dependent manner either by the NK1R antagonist (netupitant, i.p.); or the L-type Ca^{2+} channel (LTCC) blocker (nifedipine, s.c.). Here we examined the downstream intracellular signaling effectors in the shrew brainstem. Using immunohistochemistry performed on brainstem slices, substance P immunoreactivity was increased in the brainstem emetic nucleus, dorsal motor nucleus of the vagus, 15 min following GR73632 administration (5 mg/kg, i.p.), but not by its 1 mg/kg non-emetic dose. Moreover, we performed Western blot on brainstems collected respectively at 0, 5, 10, 15, 20, 25, 30 min after GR73632 challenge (5 mg/kg, i.p.). GR73632 caused time-dependent increases in effector phosphorylation, such as Ca^{2+}/calmodulin kinase IIα (CaMKIIα) and extracellular signal-regulated protein kinase1/2 (ERK1/2). Netupitant or nifedipine pretreatment completely abrogated the evoked phosphorylations. Additionally, we analyzed the contribution of intracellular Ca^{2+} release channels, inositol trisphosphate receptor (IP3R) and ryanodine receptor (RyR) on the basis of intracellular Ca^{2+} elevation in response to NK1R activation by substance P and GR73632 in vitro cultured neurons as reported previously. As a result, the IP3R inhibitor 2-APB pretreatment achieved a significant reduction of NK1R-mediated emesis, however the RyR inhibitor dantrolene did not. Our present studies demonstrate GR73632-evoked emesis occurs in a Ca^{2+}-dependent manner following NK1R activation involving IP3R which is in agreement with Miyano et al. (2010)'s observation that NK1R stimulation by GR73632 in spinal astrocytes induces Ca^{2+} release through IP3R. These results extend what we have previously observed for 5-HT3Rs, suggesting an important role for the brainstem Ca^{2+}-CaMKII/ERK interplay in the mediation of NK1R-mediated emesis in least shrews. Future effort will focus on studies to evaluate whether such mediators are acting individually or in a sequential manner.

Disclosures: W. Zhong: None. **S. Chebolu:** None. **N.A. Darmani:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.07/QQ10

Topic: F.10. Food Intake and Energy Balance

Title: Elucidating the central anorexigenic mechanism of corticotropin releasing factor in chickens (Gallus gallus domesticus)

Authors: *J. WANG, E. R. GILBERT, M. A. CLINE
Dept. of Animal and Poultry Sci., Virginia Tech., Blacksburg, VA

Abstract: Central administration of corticotropin-releasing factor (CRF), a 41-amino acid peptide, is associated with anorexigenic effects across various species, such as rodents and chickens. The hypothalamus is a primary site for regulation of energy intake and expenditure and contains several important appetite-associated nuclei. To date, the central mechanism of the potent anorexigenic effect of CRF is still unclear. Hence, the primary objective of the current study was to identify hypothalamic regions and neurotransmitters that mediate CRF-induced anorexic effects in chickens. At 1 h post-intracerebroventricular (ICV) injection, there were more c-Fos immunoreactive cells in the arcuate nucleus (ARC), dorsomedial hypothalamic nucleus (DMN), ventromedial nucleus of the hypothalamus (VMH), and the paraventricular nucleus (PVN) of 0.02 nmol CRF-injected than vehicle-injected chicks. Neuropeptide Y receptor sub-type 1 (NPYR1) mRNA was greater in the hypothalamus of chicks injected with CRF than those injected with vehicle. In the ARC, mRNA expression of orexigenic factors including neuropeptide Y (NPY), neuropeptide Y receptor sub-types 1 (NPYR1) and agouti-related peptide (AgRP) and the anorexigenic factors such as CRF, CRF receptor sub-types 1 (CRFR1) were decreased and increased, respectively. In the DMN, NPYR1 mRNA decreased in CRF-treated chicks. The exogenous administration of CRF increased CRF receptor sub-type 2 (CRFR2) and mesotocin (MT) mRNA in the PVN and VMH, respectively. In conclusion, our results suggest that the anorexigenic effects of CRF in the chicken are primarily influenced by overriding anorectic tone originated from the competitive interaction between CRF and NPY in the ARC and that the neural pathways connected the ARC with other appetite-associated nuclei are essential for mediating such anorectic tone within the hypothalamus.

Disclosures: J. Wang: None. **E.R. Gilbert:** None. **M.A. Cline:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.08/QQ11

Topic: F.10. Food Intake and Energy Balance

Title: Central neuropeptide FF injection is associated with anorexigenic effects in Japanese quail

Authors: *A. L. LOGAN, E. R. GILBERT, M. A. CLINE
Animal and Poultry Sci., Virginia Polytechnic Inst. and State Universit, Blacksburg, VA

Abstract: Central neuropeptide FF (NPFF) administration was associated with anorexigenic effects in domesticated broiler chickens (*Gallus gallus*). The purpose of the present experiment was the evaluate the effects of NPFF on feeding behavior in a less artificially-selected avian species, the Japanese quail (*Coturnix japonica*). In Experiment 1, 7 day-old Japanese quail were intracerebroventricularly injected with 0 (vehicle), 8, 16, or 32 nmol NPFF and food and water intake were quantified over a three-hour period. During the first thirty min post-injection, food intake was decreased in the 32 nmol NPFF-injected compared to the vehicle-injected group, while water intake was not affected. In Experiment 2, we conducted a comprehensive behavior analysis following central injection of 0 or 32 nmol NPFF. In NPFF-treated quail, the number of water pecks, distance moved, and steps taken significantly decreased, while the time spent in deep rest significantly increased, compared to vehicle-injected quail. In Experiment 3, we collected the hypothalamus at 1 h post-injection of 0 or 32 nmol NPFF and measured mRNA abundance of appetite-associated factors via real time PCR. NPFF injection was associated with increased expression of melanocortin receptor sub-type 3 (MC3R) mRNA and decreased abundance of neuropeptide Y receptor 1 (NPYR1) and prodynorphin (PDYN) mRNA, compared to vehicle injection. In sum, there are threshold differences in food intake and behavior between quail and chicken in response to central injection of NPFF. Additionally, the hypothalamic mechanism mediating NPFF-induced satiety in quail may involve MC3R, NPYR1 and PDYN.

Disclosures: A.L. Logan: None. **E.R. Gilbert:** None. **M.A. Cline:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.09/QQ12

Topic: F.10. Food Intake and Energy Balance

Support: Intramural Program, NIH

National Science and Engineering Research Council of Canada (NSERC) Discovery Grant (RGPIN-2016-06023), a Canadian Foundation for Innovation Grant (CFI Project 35297), University of Toronto Mississauga start-up funds

Title: Neuropeptigeric control of feeding in Trichoplax, an animal lacking synapses

Authors: C. L. SMITH[1], *T. S. REESE[2], A. SENATORE[3]
[1]NINDS, [2]NINDS - Lab. of Neurobio., NIH, Bethesda, MD; [3]Neurosci. Inst., Univ. of Toronto, Mississauga, ON, Canada

Abstract: *Trichoplax adhaerens* is a flat, millimeter-sized marine animal that adheres to surfaces and grazes on algae, displaying a repertoire of different feeding behaviors despite the apparent

absence of a true nervous system with electrical or chemical synapses. It glides along surfaces to find food, propelled by beating cilia on cells at its ventral surface, and pauses during feeding by arresting ciliary beating. We find that when endomorphin-like peptides are applied to an animal, ciliary beating is arrested, mimicking natural feeding-pauses. Antibodies against these neuropeptides label cells that express the neurosecretory proteins and voltage gated calcium channels implicated in regulated secretion. These cells are embedded in the ventral epithelium, where they comprise only four percent of total cells, and are concentrated around the edge of the animal. Each bears a cilium likely to be chemosensory and used to detect algae. *Trichoplax* pausing during feeding or spontaneously in the absence of food often induce their neighbors to pause as well, even neighbors not in direct contact. Pausing-behavior propagates from animal to animal across distances much greater than the signal that diffuses from just one animal, so we presume that the peptides secreted from one animal elicit additional secretion from nearby animals. Signal amplification by peptide-induced peptide secretion explains how a small number of sensory secretory cells lacking processes and synapses can evoke a wave of peptide secretion across the entire animal to globally arrest ciliary beating and allow pausing during feeding.

Disclosures: C.L. Smith: None. **T.S. Reese:** None. **A. Senatore:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.10/QQ13

Topic: F.10. Food Intake and Energy Balance

Title: Alpha-melanocyte stimulating hormone-induced anorexia in Japanese quail(Corturnix japonica) likely involves the ventromedial hypothalamus and paraventricular nucleus of the hypothalamus

Authors: *M. O'DONNELL[1], E. GILBERT[1], M. A. CLINE[2]
[2]Animal and Poultry Sci., [1]Virginia Tech., Blacksburg, VA

Abstract: Alpha-melanocyte stimulating hormone (α-MSH) reduces food intake in birds and mammals. The objective of this experiment was to determine effects of α-MSH on food and water intake, and hypothalamic c-Fos immunoreactivity and appetite-associated factor mRNA in Japanese quail (*Coturnix japonica*), a gallinaceous species that has not undergone the same artificial selection for growth-related traits as the chicken. At 7 days post-hatch, 3-hour-fasted quail were intracerebroventricularly (ICV) injected with 0 (vehicle), 0.5, 5, or 50 pmol of α-MSH and food and water intake were recorded at 30 minute intervals for 180 minutes. In the second and third experiment, quail were injected with 50 pmol α-MSH and hypothalami were collected at 1 hour to determine c-Fos immunoreactivity and mRNA abundance, respectively. At 30 minutes, chicks injected with 5 or 50 pmol of α-MSH ate and drank less than vehicle-injected

chicks. Chicks injected with 50 pmol ate less for the entire duration of the experiment and drank less than vehicle-injected chicks for 120 minutes post-injection. Corticotropin-releasing factor mRNA was greater in male than female α-MSH-injected chicks. Expression of agouti-related peptide and DOPA decarboxylase were greater in vehicle- than α-MSH-injected quail, whereas melanocortin receptor 4 (MC4R) mRNA was greater in α-MSH- than vehicle-injected birds. Alpha-MSH injection was associated with more c-Fos immunoreactive cells in the ventromedial hypothalamus (VMH) and paraventricular nucleus (PVN) of the hypothalamus. Results suggest that the anorexigenic effect of α-MSH is conserved among avians and that effects are mediated via the VMH and PVN and involve MC4R.

Disclosures: M. O'Donnell: None. **E. Gilbert:** None. **M.A. Cline:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.11/QQ14

Topic: F.10. Food Intake and Energy Balance

Support: Georgia State University

Title: The role of ventral tegmental area melanocortin 3 receptor-expressing neurons in the regulation of feeding and body weight

Authors: *A. I. DUNIGAN, A. G. ROSEBERRY
Biol., Georgia State Univ., Atlanta, GA

Abstract: The melanocortin and the mesolimbic dopamine (DA) systems are both intricately involved in the regulation of feeding and body weight. The melanocortin system, which is comprised of proopiomelanocortin and agouti-related protein neurons, the neurotransmitters they release, and their receptors, including the MC3R and MC4R melanocortin receptors, is well known for its role in controlling normal, baseline, need-based feeding, whereas the mesolimbic DA system, which is comprised of the DA neurons of the ventral tegmental area (VTA) and their afferent and efferent projections, is the primary neural circuit controlling reward and motivated behavior, including the rewarding and motivational qualities of food. Increasing evidence indicates that the melanocortin system can act on dopamine circuits to control feeding. For example, intra-VTA injection of MC3R/MC4R agonists increases DA release and decreases both need-based and reward-based feeding. The mechanisms underlying these behavioral effects, including the identity of the melanocortin receptor subtypes mediating these effects, and how alterations in VTA neuron activity lead to the observed changes in feeding are unknown, however. In these studies we used transgenic mice expressing cre recombinase in MC3R-expressing neurons (MC3R-cre mice) to examine the role of VTA MC3R neurons in the control

of feeding and body weight. Viral tracing was used to map the efferent projections of the VTA MC3R neurons and optogenetics coupled with brain slice electrophysiology were used to determine the identity of the neurotransmitters released by VTA MC3R axons at these targets. We have also used Designer Receptors Exclusively Activated by Designer Drugs to test the effects of activation and inhibition of VTA MC3R neurons on feeding and body weight. Our data indicate that VTA MC3R neurons project to a restricted set of efferent target regions and release multiple neurotransmitters in the different efferent targets. Furthermore, activation of VTA MC3R neurons appears to acutely decrease feeding and body weight. These studies advance our understanding of how the melanocortin and mesolimbic dopamine systems interact to control feeding and body weight through the regulation of the activity of VTA MC3R neurons.

Disclosures: A.I. Dunigan: None. **A.G. Roseberry:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.12/QQ15

Topic: F.10. Food Intake and Energy Balance

Support: USDA Grant 422471

Title: Peripheral neuropeptide Y differentially influences adipogenesis and lipolysis in chicks from lines selected for low or high body weight

Authors: *Y. XIAO, L. LIU, G. WANG, S. L. SHIPP, P. B. SIEGEL, M. A. CLINE, E. R. GILBERT
Dept. of Animal and Poultry Sci., Virginia Polytechnic Inst. and State Univ., Blacksburg, VA

Abstract: Neuropeptide Y (NPY) stimulates appetite and promotes lipid deposition in the adipose tissue. We previously demonstrated a differential sensitivity in the food intake response to central NPY in chicks from lines selected for low (LWS) or high (HWS) body weight. The LWS are relatively lean and hypophagic whereas HWS are all compulsive eaters that become obese. The purpose of this study was to determine whether such differences exist between the lines in adipose tissue metabolism after peripheral NPY administration. At 5 days post-hatch, LWS and HWS chicks were intraperitoneally injected with 0 (vehicle), 60, or 120 μg/kg BW NPY. Subcutaneous adipose tissue and plasma were collected at 1, 3, 6, 12, and 24 h post-treatment (n=12 per treatment per time point). Peripheral NPY injection increased food intake in both lines. In response to NPY injection, glycerol-3-phosphate dehydrogenase (G3PDH) activity at 1 and 3 h was increased and was greater in HWS than LWS, whereas plasma non-esterified fatty acids (NEFAs) were reduced at 1 and 12 h and were lower in HWS than LWS. At 1 h, peroxisome proliferator-activated receptor gamma (PPARγ), CCAAT/enhancer binding protein α

(C/EBPα), and microsomal triglyceride transfer protein (MTTP) mRNAs were reduced in NPY-injected chicks whereas NPY receptor 1 (NPYR1) mRNA was increased, relative to vehicle-injected chicks. Expression of stearoyl-CoA desaturase (SCD1) was increased by NPY at 1 h in HWS but not LWS. PPARγ (3 and 6 h), C/EBPβ (3 h), C/EBPα (6 h) and NPYR1 and NPYR2 (24 h) mRNAs were greater in NPY- than vehicle-injected chicks. At several times, genes encoding factors associated with lipolysis (adipose triglyceride lipase, perilipin 1, NPYR1), and adipogenesis (MTTP and NPYR2) were more highly expressed in LWS than HWS, while expression of SCD1, glycerol-3-phosphate acyltransferase 3 and lipoprotein lipase, which encode enzymes that are associated with lipid synthesis and deposition, were greater in HWS than LWS. In conclusion, NPY promotes adipogenesis and inhibits lipolysis in chicks, with line differences indicative of greater rates of lipolysis in the lean LWS and adipogenesis in obese HWS chicks.

Disclosures: Y. Xiao: None. **L. Liu:** None. **G. Wang:** None. **S.L. Shipp:** None. **P.B. Siegel:** None. **M.A. Cline:** None. **E.R. Gilbert:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.13/QQ16

Topic: F.10. Food Intake and Energy Balance

Support: FAPESP 2016/02748-0

FAPESP 2014/13489-0

FAPESP 2010/52068-0

CAPES COFECUB 848/15

CNPQ Researcher Grant

Title: Morphological and neurochemical characterization of the melanin-concentrating hormone [MCH] peptidergic system in the CNS of the mouse

Authors: *G. B. DINIZ[1], P. M. CHERUBINI[1], J. C. G. DUARTE[1], J. G. P. FERREIRA[1], D. S. BATAGELLO[1], L. V. SITA[1], J. C. BITTENCOURT[1,2]
[1]Dept. of Anat., Inst. of Biomed. Sci., Univ. of São Paulo, Sao Paulo, Brazil; [2]Ctr. for Neurosci. and Behavior, Inst. of Psychology, Univ. of São Paulo, São Paulo, Brazil

Abstract: The *Pmch* is a highly-conserved gene in vertebrate species. It encodes three peptides, the melanin-concentrating hormone [MCH], Neuropeptide E-I [NEI] and Neuropeptide G-E [NGE]. MCH has been implicated in several biological processes, including the coordination of

motivated behaviors, autonomic modulation, memory, attention and sensory integration, while NEI has been associated to thyroid modulation and locomotor activity. Although the morphological and hodological aspects of this peptidergic system have been thoroughly examined in rats, there is a paucity of systematic evaluations of this system in mice. This has led to an information mismatch in the literature, as functional data obtained from mice are commonly interpreted using anatomical data from rats, without any confirmation of the transposability of such data. To provide accurate morphological and neurochemical data regarding the MCH peptidergic system in mice, we examined the distribution of MCH and NEI in male and female mice, including animals in all stages of the reproductive cycle, using immunohistochemistry tools. We additionally examined another peptidergic system that spatially correlates to MCH, the orexin system [ORX], to obtain comparative reference. Our results indicate that, although numerous morphological aspects are similar between rats and mice regarding the MCH peptidergic system, important differences were found regarding structures linked to endocrine modulation, in addition to a dampened response to the reproductive cycle in mice, suggesting a decreased participation of MCH and NEI in these functions. Furthermore, the comparison of our results to other mammals suggest that MCH conserved its diencephalic location, but varies between species in terms of dorso-ventral, latero-medial and rostro-caudal distributions. Finally, we propose that MCH and ORX are organized into two chemical domains in the lateral hypothalamus, the LHA shell and the LHA core that may have important implications for the intercommunication between these two neuronal populations.

Disclosures: G.B. Diniz: None. **P.M. Cherubini:** None. **J.C.G. Duarte:** None. **J.G.P. Ferreira:** None. **D.S. Batagello:** None. **L.V. Sita:** None. **J.C. Bittencourt:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.14/QQ17

Topic: F.10. Food Intake and Energy Balance

Support: NIH R01 HD69702

Title: Leptin promotes growth and bone formation via the PI3Kα signalling pathway

Authors: *D. GARCIA GALIANO[1], S. J. ALLEN[2], K. M. KOZLOFF[3], C. F. ELIAS[2]
[2]Mol. and Integrative Physiol., [3]Biomed. Engin., [1]Univ. of Michigan, Ann Arbor, MI

Abstract: Bone metabolism is intimately related to energy balance and fat reserves, and the brain is directly implicated in this regulation. Leptin is secreted peripherally to promote the bone formation via modulation of GHRH-GH-Igf-1 axis. However, more detailed work is required to understand the molecular mechanisms associated with leptin action. In this sense,

phosphatidylinositol 3-kinase (PI3K) pathway plays an essential role in long-term energy homeostasis, mediating leptin and growth factors actions in endocrine modulation. Using mouse models with specific deletion of the PI3K p110α catalytic subunit (PI3Kα) in leptin receptor (LR)-expressing cells ($LR^{\Delta\alpha}$), we observed a decreased body weight and snout-anus distance in $LR^{\Delta\alpha}$ mice respect to floxed controls at postnatal day of 60 (P60). Micro-computed tomography detected a decrease in bone mineral density at trabecular and reduced cortical area in femur of $LR^{\Delta\alpha}$ females (n=6/genotype). Additionally, a significant increase in AgRP transcript in the arcuate nucleus and fiber projections into hypothalamic paraventricular nucleus were observed in $LR^{\Delta\alpha}$ mice (n=6/genotype). Analysis of gene expression at different levels of the growth hormone (GH) axis did not detect differences in hypothalamic GHRH or SST mRNA expression between genotypes at P60. However, a significant reduction in hepatic GH-R and Igf-1 mRNA expression was detected in adult $LR^{\Delta\alpha}$ mice. Reduced growth rate was only detected in sexually mature $LR^{\Delta\alpha}$ females, with no differences in pubertal (P40) stage, suggesting an effect of sex steroids on growth via PI3Kα pathway. Finally, we explored if leptin supplementation can affect the growth rate in pubertal mice. For this, peripubertal females of both genotypes were daily injected with leptin from P35-P55. Leptin supplementation in control females decreased cumulative weight gain but body length was increased at P60. Notwithstanding, leptin treatment had an enhanced effect on weight loss but no changes in growth rate were detected in $LR^{\Delta\alpha}$ females. These data suggest that $LR^{\Delta\alpha}$ mice are unresponsive to leptin's effect on growth rate. Collectively, our findings indicate that the PI3Kα downstream of leptin signaling is required for normal progression of growth and bone formation at pubertal stages.

Disclosures: D. Garcia Galiano: None. **S.J. Allen:** None. **K.M. Kozloff:** None. **C.F. Elias:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.15/QQ18

Topic: F.10. Food Intake and Energy Balance

Support: CIHR RNL-132870

RDC 5404.1171.102

Title: Prostaglandin E_2 activates melanin-concentrating hormone neurons in diet-induced obesity

Authors: L. FANG, V. LINEHAN, *M. HIRASAWA
Div. of Biomed. Sci., Mem. Univ., St John's, NL, Canada

Abstract: Diet-induced obesity is associated with low-grade chronic inflammation of the hypothalamus, a key brain region for energy homeostasis. However, it remains unclear how this neuroinflammation influences appetite-related neurons. Melanin-concentrating hormone (MCH) neurons are one of the hypothalamic cell groups that promote appetite and weight gain. Therefore, any influence of neuroinflammatory factors on MCH neurons may modulate energy balance. Prostaglandin E_2 (PGE_2) is an important mediator of hypothalamic inflammation under disease states and may also play a role in obesity-related inflammation. Thus, we asked whether PGE_2 modulates the activity of MCH neurons in diet-induced obesity. To answer this question, 3-week old male Sprague Dawley rats were fed *ad libitum* with palatable high-fat diet (HFD) or control chow for >11 weeks. Following the feeding period rats were sacrificed and whole-cell patch clamp recordings were performed on acute brain slices. We found that the resting membrane potential of MCH neurons was depolarized in the HFD group compared to chow controls. This excitatory effect of HFD was mimicked by incubating chow control slices with a low concentration of PGE_2, which was blocked by the EP2 receptor-specific antagonist PF 04418948, but not by antagonists for other subtypes of PGE_2 receptors (EP1, EP3, EP4). HFD-induced depolarization was also reversed by the EP2 receptor antagonist, suggesting a role of PGE_2 in the HFD effect. In support of this, inhibiting PGE_2 production in the HFD brain slice by the COX inhibitor acetaminophen reversed the depolarization of MCH neurons. Using COX-1 and COX-2 specific inhibitors SC-560 and SC-236 respectively, we further determined that COX-2 is responsible for the PGE_2 production in the hypothalamus of HFD-fed animals. In conclusion, our study shows that HFD-induced activation of MCH neurons is mediated by the COX-2/PGE_2 pathway. Given the known role of MCH in promoting positive energy balance, this mechanism may be at least partially responsible for diet-induced obesity. Therefore, elucidating the mechanism behind MCH neuron activation in obese conditions will provide insight into potential new therapies for obesity.

Disclosures: L. Fang: None. **V. Linehan:** None. **M. Hirasawa:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.16/QQ19

Topic: F.10. Food Intake and Energy Balance

Support: NIH Grant DA040782

Title: Neural adaptation in the LH area in mice exposed to chronic high-fat diet

Authors: F. HANG[1], Z.-W. LIU[2], Y. TAN[3], *X.-B. GAO[4]
[1]Guangxi Reproductive Med. Res. Ctr., First Affiliated Hosp. of Guangxi Med. Univ., Nanning, Guangxi, China; [2]Comparative Med., Yale university Sch. of Med., New Haven, CT; [3]The

Second Affiliated Hosp. of Chongqing Med. Univ., Chongqing, China; [4]Yale Univ. Sch. Med., New Haven, CT

Abstract: Drug addiction is a chronic and relapsing disease resulting from repeated exposure to the drugs of abuse. The sensitivity to drugs of abuse is regulated by many physiological and pathological conditions, particularly the metabolic status of animals. Food restriction increases and over-nutrition decreases the sensitivity to drugs of abuse. However, it is still elusive how the brain circuitry regulating the metabolic status interacts with the reward circuitry. The perifonical/lateral hypothalamus is a critical brain area to regulate both energy homeostasis and food/drug reward. A selective group of neurons exclusively synthesizing the neuropeptide hypocretin (Hcrt, also called orexin) affect food intake and play a prominent role in food award and drug addiction. In this study, by using electrophysiological, immunocytochemical and behavioral methods we examined changes in basic synaptic properties in Hcrt cells and responses of Hcrt neurons to cocaine exposure and acute stress (a cue triggering relapse of cocaine use) in control and diet-induced obesity (DIO) mice. C57/B6 mice at an age of 4 weeks old were divided into two groups: control, fed with a normal diet (ND) for 10-12 weeks; DIO group, fed with a high fat diet (45% calories from fat, Research Diets Inc.) for 10-12 weeks. Whole-cell patch clamp recording was performed in GFP-labeled Hcrt cells in brain slices from both groups. The frequency of mEPSCs was 218.8 ± 39.9 /min (n=12) and 137.4 ± 28.7/min (n=12) in Hcrt cells in ND and DIO mice, respectively. The difference between these two groups is not statistically significant but the trend is obvious (p=0.11, t test). The frequency of mIPSCs was 70.4 ± 13.4 /min (n=12) in Hcrt cells in ND and 68.4 ± 22.5 /min (n=12) in Hcrt cells in DIO mice (p>0.05, t test). In addition, we have examined evoked excitatory postsynaptic currents in Hcrt cells in ND and DIO mice. Under a train of stimulations (100 Hz, 20 pulse) to Hcrt cells, the evoked EPSCs decayed rapidly (within 20 pulses) in these cells in ND mice but did not decay as fast in the DIO group. Since the amplitudes of evoked EPSCs induced by these stimuli decay as the readily releasable pool (RRP) of vesicles in presynaptic terminals are depleted by the rapid, repetitive stimulations, this result and the decrease in the frequency of mEPSCs suggest a possible de-potentiation of presynaptic efficacy in Hcrt neurons in DIO mice. Lastly, our data indicate that the responses of Hcrt cells to cocaine treatments and acute stress were altered as well. In conclusion, our preliminary tests suggest a re-programed Hcrt system by high-fat diet may be responsible for altered behaviors relevant to cocaine reward/abuse in animals.

Disclosures: **F. Hang:** None. **Z. Liu:** None. **Y. Tan:** None. **X. Gao:** None.

Poster

416. Neuropeptide Regulators

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 416.17/QQ20

Topic: F.10. Food Intake and Energy Balance

Title: Anorexigenic effects of substance P in Coturnix japonica

Authors: *C. BUENAVENTURA[1], M. A. CLINE[2], E. R. GILBERT[3]
[2]Animal and Poultry Sci., [1]Virginia Tech., Blacksburg, VA; [3]Animal Sci., Virginia Polytechnic Inst. and State Univ., Blacksburg, VA

Abstract: When substance P (SP), an 11 amino acid-residue tachykinin that is produced centrally and peripherally, is centrally administered, there is reduced food intake although the mechanism mediating this effect is poorly understood. Hence, the objective of this project was to elucidate the central mechanism associated with reduced food intake using Japanese quail (*Coturnix japonica*) as a model. In Experiment 1, 7 day-old quail were intracerebroventricularly injected with 0, 0.25, 0.5 or 1.0 nmol SP and food and water intake were quantified for 180 min. On a cumulative basis, quail that received 0.5 and 1.0 nmol SP reduced food intake during the entire observation period (50% reduction compared to controls at 30 min post-injection) while those administered 0.25 nmol were not different from vehicle-injected chicks. On a non-cumulative basis, food intake in the 0.5 and 1.0 nmol-injected groups was different from the vehicle-injected group at only 30 min post-injection. Water intake was not affected. In Experiment 2, whole hypothalamus was isolated from injected chicks and real-time PCR performed to measure mRNA abundance of several appetite-associated factors. Quail injected with SP had reduced agouti-related peptide (AgRP) mRNA compared to quail injected with vehicle. In Experiment 3, a comprehensive behavior analysis was performed. Quail that received SP had decreased feeding pecks and reduced locomotion compared with quail that received vehicle. In sum, SP induces a potent anorexia following central injection in quail that coincides with reduced hypothalamic AgRP expression. The effect on food intake may be primary because SP did not affect behaviors that would be competitive with food intake.

Disclosures: C. Buenaventura: None. **M.A. Cline:** None. **E.R. Gilbert:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.01/QQ21

Topic: G.01. Appetitive and Aversive Learning

Support: NIH Grant MH107239

Title: Gamma oscillations in the basolateral amygdala and prefrontal cortex support the emotional modulation of memory consolidation

Authors: *V. KANTA, D. PARÉ, D. B. HEADLEY
Rutgers, the State Univ. of New Jersey, Newark, NJ

Abstract: Recently formed memories are labile, but with time can strengthen and become permanent through a process of consolidation. Emotional arousal enhances consolidation, resulting in memories that are vivid and long lasting. This effect is mediated by the basolateral amygdala (BLA) and its strong projections to both the hippocampus and neocortex. While there has been extensive investigation into the electrophysiological mechanisms of consolidation between the hippocampus and neocortex, comparatively little is known about the BLA's role. What is known is that the BLA generates gamma oscillations during emotional arousal, and artificially driving a gamma rhythm in the BLA boosts the consolidation of aversive memories. Thus, gamma oscillations constitute a promising candidate mechanism for mediating the enhancing effects of emotional arousal on memory consolidation. In this study, we examine how the consolidation of emotional experiences affects the interactions between the BLA, ventral hippocampus, and medial prefrontal cortex (mPFC), a neocortical area linked to memory consolidation. In particular, we study how gamma-related activity in these areas is affected during consolidation of both aversive and positively motivated spatial memories. The behavioral tasks we used are inhibitory avoidance and the holeboard foraging task. Preliminary results show that gamma power and coherence between BLA and mPFC (but not ventral hippocampus) increased immediately after training on both tasks. Crucially, its strength correlates with subsequent retention. To establish whether changes in gamma are causally related to memory consolidation, we have developed a closed-loop real-time optogenetic control system that can reliably detect oscillatory bursts in the BLA with frequency and phase specificity. Using this system, we intend to modulate gamma oscillations during the post-training consolidation period. Overall, our results indicate that the coordination of gamma oscillations between the BLA and mPFC supports consolidation of emotional memories.

Disclosures: V. Kanta: None. **D. Paré:** None. **D.B. Headley:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.02/QQ22

Topic: G.01. Appetitive and Aversive Learning

Support: NIH Grant R01MH098348

Title: Stress-induced changes in effective connectivity during the emotional response to threat

Authors: *A. M. GOODMAN[1], M. D. WHEELOCK[2], N. G. HARNETT[1], D. R. HURST[1], T. R. OREM[1], G. DESHPANDE[3], S. MRUG[1], D. C. KNIGHT[1]

[1]Psychology, Univ. of Alabama at Birmingham, Birmingham, AL; [2]Psychiatry, Washington Univ., St. Louis, MO; [3]Electrical and Computer Engin., Auburn Univ. MRI Res. Ctr., Auburn, AL

Abstract: Stress exposure can disrupt emotional learning and regulation processes. However, the neurobiological processes that mediate stress-related dysfunction that impacts learning and memory is not well understood. Studies from our laboratory have shown that conditioned diminution of the emotional response to threat (reduced emotional response to predictable vs unpredictable threat) is a regulatory process supported by a prefrontal cortex (PFC)-hippocampus-amygdala neural circuit during fear conditioning. The current study investigated the hypothesis that stress disrupts the functional connectivity of the PFC-hippocampus-amygdala circuit, and in turn, the dysfunction of this circuit disrupts emotion regulation. 120 right-handed volunteers completed a variant of the Montreal Imaging Stress Task (MIST) followed (20 minutes later) by a Pavlovian fear conditioning procedure during fMRI. Self-reported stress and psychophysiological responses to the MIST were used to assess stress reactivity. Psychophysiological measures and fMRI signal during fear conditioning were used to assess autonomic responses and neural activity underlying conditioned diminution of the emotional response to threat. In order to find task-specific connectivity during the fear conditioning, Granger Causality (GC) values were populated into two different samples based on the predictable (CS+UCS; UCS that follows the CS+) and unpredictable (UCS alone) threat presentations. Analyses of predictable and unpredictable trials of the UCS revealed two main hub ROIs: the dorsolateral PFC (dlPFC) and insula. Presentations of the predictable threat (i.e., CS+UCS) showed greater connectivity than unpredictable threat (i.e., UCS alone) from the dlPFC to dorsomedial PFC (dmPFC), ventromedial PFC (vmPFC), dlPFC, posterior cingulate cortext (PCC), amygdala, and inferior parietal lobule (IPL). Presentations of the CS+UCS showed connectivity from the dlPFC to the insula, PCC, vmPFC, IPL, and amygdala, and from the insula to the dlPFC, PCC, and amygdala. Connectivity during the unpredictable threat was observed from the dlPFC to the vmPFC, dmPFC, dlPFC, insula, PCC, IPL, and amygdala. Stress reactivity (self-reported) did not vary with connectivity during the predicable threat. In contrast, stress reactivity varied with connectivity during UCS alone trials from the insula to the IPL. These findings suggest that the left dlPFC was significantly more involved in top-down regulation of other brain regions during CS+UCS trials compared to UCS alone. Increased connectivity between the insula and IPL may play a critical role in stress-induced changes in the emotional response to threat.

Disclosures: **A.M. Goodman:** None. **M.D. Wheelock:** None. **N.G. Harnett:** None. **D.R. Hurst:** None. **T.R. Orem:** None. **G. Deshpande:** None. **S. Mrug:** None. **D.C. Knight:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.03/RR1

Topic: G.01. Appetitive and Aversive Learning

Support: ASU College of Liberal Arts and Sciences

Title: How do rest periods following the end of chronic stress impact fear conditioning?

Authors: *J. M. JUDD, V. SHAH, A. FLEGENHEIMER, B. LE, F. SANABRIA, C. D. CONRAD
Psychology, Arizona State Univ., Tempe, AZ

Abstract: In fear conditioning paradigms, chronic stress enhances fear learning (Conrad *et al.*, 1999), attenuates extinction learning (Izquierdo *et al*., 2006), and impairs recall of extinction (Baran *et al.*, 2009). Chronic stress increases anxiety-like behavior, which remains elevated even after a no-stress period (i.e., rest) prior to the start of behavioral testing (Vyas *et al*., 2004). In other tasks, the effects of chronic stress seem to disappear following a rest period (Conrad *et al.*, 2016). Whether a rest period on fear conditioning and extinction would lead to persistent effects or dissipate is untested. Male Sprague-Dawley rats were chronically stressed by wire mesh restraint (6hr/d/21) or assigned to a non-stressed control group (CON). Fear conditioning occurred within a day (IMM), 3 weeks (REST3), or 6 weeks (REST6) after chronic stress ended. Prior to the start of fear conditioning, rats were acclimated to the training and testing contexts for 10mins/day 3days/context to attenuate the likelihood of chronic stress potentiating fear learning during the training session. Fear conditioning involved the presentation of 3 tone (20sec, 2kHz, 75db)-footshock (1sec, 0.8mA) pairings. Extinction took place over the next two days in a different context than during training and consisted of 15 tone presentations (no footshocks). The duration of freezing to each tone and context (20-secs immediately prior to each tone), and the difference between both durations (difference score) were quantified. During fear conditioning, REST6 acquired the freezing response slower than the other groups; all groups showed similar and elevated freezing by the third tone presentation. In the first extinction session, rats displayed minimal freezing to the context before the first tone presentation, but differences emerged in subsequent context assessments. IMM froze significantly more to context than REST3 and REST6. All groups froze similarly to tones during extinction 1. The difference score for IMM was at zero, indicating similar freezing to tone and context, whereas REST3 and REST6 exhibited positive difference scores, indicating more freezing to tone than to context. CON difference scores were higher than IMM but lower than REST3 and REST6. No significant differences in freezing were detected during the second extinction session and spontaneous recovery. These results suggest that a rest period following the end of chronic stress may lead to

optimal fear conditioning and extinction compared to fear conditioning soon after chronic stress has ended. Establishing whether the IMM performance during extinction reflects generalization is underway.

Disclosures: J.M. Judd: None. **V. Shah:** None. **A. Flegenheimer:** None. **B. Le:** None. **F. Sanabria:** None. **C.D. Conrad:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.04/RR2

Topic: G.01. Appetitive and Aversive Learning

Support: ARC Grant DP160100004

Title: Midbrain dopamine neurons regulate model-based, not model-free, aversive learning

Authors: *C. Y. PENG, P. JEAN-RICHARD DIT BRESSEL, G. P. MCNALLY
Psychology, Univ. of New South Wales - Kensington Campus, Sydney, Australia

Abstract: Whereas dopamine is well-known for mediating reward learning, its role in aversive learning and threat-related attention is less well understood. Here we used optogenetics and transgenic TH::Cre rats to investigate the causal role of Ventral Tegmental Area (VTA) Tyrosine hydroxylase (TH) neurons in aversive learning under experimental designs enabling concurrent assessment of model-free (simple Pavlovian conditioning) and model-based (punishment) learning. First, we show in simple model-free Pavlovian fear conditioning that optogenetic silencing of VTA TH+ neurons during the conditioned stimulus or unconditioned stimulus has no effect on fear learning, across different asymptotic levels of learning and across differences in strength of positive prediction errors. Then, we show that optogenetic inhibition of VTA TH+ neurons, in the same animals, is sufficient to punish instrumental responding for a food reward. To directly compare the role of VTA TH neurons in these two forms of aversive learning we developed a novel behavioural paradigm that allowed concurrent assessment of model-free aversive learning (fear) and model-based aversive learning (punishment) under identical stimulus and shock contingencies. Using this paradigm we show that optogenetic inhibition of VTA TH neurons augments aversive model-based learning without affecting aversive model-free learning acquired under matched conditions. These findings show that midbrain TH neurons regulate model-based, not model-free, aversive learning.

Disclosures: C.Y. Peng: None. **P. Jean-Richard dit Bressel:** None. **G.P. McNally:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.05/RR3

Topic: G.01. Appetitive and Aversive Learning

Support: NIH Grant NS48156

Title: Safety-signal learning in Hermissenda produced by conditioned inhibition training

Authors: *J. FARLEY
Indiana Univ. Bloomington, Bloomington, IN

Abstract: Previously, we found that conditioned inhibition (CI) learning in *Hermissenda* (*H.c.),* produced by repeated explicitly-unpaired (EU) presentations of light (CS) and rotation (US), fixed ISI of 4.5 min, resulted in increased phototactic behavior and decreased/increased photoresponses and spike activity of ocular Type B/A photoreceptors (Britton & Farley, 1999, *J Neurosci*; Walker et al., 2010, *Front Beh Neuro*). Recent studies (Farley et al., 2017) determined the ionic conductance changes that underlie persistent EU-produced decreases in B cell light-responses and spike frequencies: increases in two somatic K^+ currents (I_A and $I_{K\text{-}Ca}$). These same two K^+ currents are suppressed by CS-US pairing-produced persistent PKC-activity, and underlie the enhanced photoresponses and excitability of B cells that comprise the molecular basis of associative memory in *H.c.* Another often-used CI/safety-signal procedure with vertebrates, more complex than EU-training, is the "Pavlovian Conditioned Inhibition" procedure (Pav CI). Here I report that *H.c.* exposed to Pav CI training show enhanced phototactic behavior, similar to EU-trained animals. Animals in the Pav CI procedure received an initial 2 training days in which a chemosensory cue (scallop extract) was paired with rotation (C+), 25X per session. This was followed by 3 additional training days in which 50 non-reinforced (no rotation) simultaneous compound presentations of the chemosensory cue and light (CL-), intended to establish L as a signal for the "absence of rotation", were added to the C+ trials. A discrimination control group (C+/L-) of animals received the same training as the Pav CI group, except that the non-reinforced light-alone trials (L-) did not include the presentation of the chemosensory cue. During post-training open-field phototactic tests, Pav CI animals clearly moved *towards* the center of the light gradient [pre-post distance scores of (mean +/- sem)]: 4.2 +/-0.8 cm, 3.7 +/- 0.7 cm (retention test days 1 & 2, respectively), similar to EU-animals. In contrast, discrimination control (C+/L-) animals showed only small (< 0.6 cm) non-significant changes in phototaxis. Additional studies showed an acquisition function for Pav CI, as well as "extinction" of Pav CI by non-reinforced light-alone trials.

Disclosures: J. Farley: None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.06/RR4

Topic: G.01. Appetitive and Aversive Learning

Support: MH086947

Title: Neurotoxic damage to orbitofrontal subregions 12 or 13 impairs startle response to fearful and safe stimuli in macaques

Authors: *L. E. MURPHY, C. WYNN, V. WERTMAN, A. M. KAZAMA, J. BACHEVALIER
Emory Univ., Atlanta, GA

Abstract: Dysfunction of the orbitofrontal cortex (OFC) plays a key role in anxiety disorders in humans, characterized by exaggerated anxiety and fear responses to relatively harmless stimuli (Milad & Rauch, 2007). PTSD patients exhibit similar characteristics, and are unable to integrate safety-signal information in the presence of fearful cues, as measured by the safety-signal (AX+/BX-) paradigm (Jovanovic et al., 2012). Previously, we showed that selective damage to OFC area 14 impairs the expression of learned fear and safety cues in macaques on the AX+/BX- task (Kazama et al. 2013). Because the OFC includes several subregions with distinct but complimentary functions, we tested here the effects of selective lesions of OFC areas 12 and 13 on the expression of fear and safety learning and conditioned inhibition. Nine adult rhesus macaques were trained to discriminate between a fear (AX+) and a safety (BX-) cue. Percent fear-potentiated startle (%FPS) measured both the rate of associating the conditioned cues (AX+, BX-) to the unconditioned stimuli (air blast; no air blast), as well as conditioned inhibition during a probe test combining the A and B cues (AB). Subjects then received either bilateral ibotenic acids lesions to OFC area 12, OFC area 13, or sham operations. Following surgery, subjects were then given the probe test with the same cues they had learned pre-surgery. Pre-surgical performance of the 3 groups were similar on both the trials to criterion during training (AX+ and BX- cues) and %FPS during the probe test (X^2=1.00, p=0.61; $F_{2,6}$=1.20, p=0.37, n^2=0.40). A RM-ANOVA comparing the %FPS to each stimulus type (AX+, BX-, AB) post-surgery revealed a trend toward Stimulus X Lesion interaction with a large effect size ($F_{4,12}$=3.00, p=0.06, n^2=0.50), indicating that sham-operated controls again showed good discrimination between the aversive and safety cues and good conditioned inhibition ($F_{2,12}$=4.35, p=0.09, n^2=0.69). By contrast, animals with damage to either OFC areas 12 or 13 did not differentiate between the previously conditioned fear and safety cues, showing impaired expression and consequently no conditioned inhibition in the post-surgery probe test (Area 12 = $F_{2,4}$=0.85, p=0.49, n^2=0.30; Area 13 = $F_{2,4}$=1.77, p=0.28, n^2=0.47). These results suggest that lesions to either OFC areas 12 or 13

significantly impair subjects' ability to retain the emotional valence of conditioned cues and, like OFC area 14, areas 12 and 13 are necessary for the expression of fear and safety learning. Thus, the interactions between OFC areas are important for emotion regulation, and may help elucidate the neural circuity of anxiety disorders.
This work was supported by grant MH086947.

Disclosures: L.E. Murphy: None. **C. Wynn:** None. **V. Wertman:** None. **A.M. Kazama:** None. **J. Bachevalier:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.07/RR5

Topic: G.01. Appetitive and Aversive Learning

Support: DA034010

MH113053

Title: Early life stress differentially impacts adult fear discrimination and increases alcohol drinking in male and female rats

Authors: *R. A. ZACHARIAS, C. ANDREANSKY, M. H. RAY, M. A. MCDANNALD
Psychology, Boston Col., Chestnut Hill, MA

Abstract: Stress during early life is a risk factor for a number of clinical disorders, such as Post-Traumatic Stress Disorder, depression and substance abuse disorders, which manifest in adulthood. Many of the disorders for which early life stress (ELS) is a risk factor are more prevalent in females, yet there is a dearth of research on how physical ELS may differentially impact males and females later in life. In order to address this significant issue, we exposed rats to ELS to investigate sex-specific behavioral effects in adulthood. Male and female Long-Evans rats were born in the laboratory and weaned on post-natal day 21. Half of the males and females underwent ELS twice daily on post-natal days 26-35, while the other half were non-exposed controls. The ELS procedure consisted of exposure to four types of adverse experiences five times each, including forced cold water swim, tail pinch, cat hair exposure, and restraint stress. Body weights were recorded in order to track physical development. Starting on post-natal day 70, all rats were maintained at 85% of their free body weight and trained to nose poke for pellets. After two days of pre-exposure to three 10-second auditory cues, all rats then underwent 16 days of Pavlovian fear discrimination during which the three different auditory cues predicted foot shock at different probabilities: 0% (safety), 25% (uncertainty), and 100% (danger). After discrimination, rats underwent 6 days of extinction, which consisted of presentations of the three

auditory cues without foot shock. At least 10 days after the conclusion of fear conditioning, all rats were given voluntary, intermittent access to 20% ethanol in 8, 24-hour sessions. Results indicated that ELS impaired physical development, resulting in decreased body weights in both male and female rats compared to their control counterparts. However, no differences in body weights were observed in adulthood. Males exposed to ELS performed similarly to controls during fear discrimination; however, females demonstrated generalized higher fear to both the safety and uncertainty cues compared to their control counterparts during discrimination and continuing into extinction. ELS rats drank more alcohol than controls in initial alcohol drinking sessions, and this effect was present in males and females. Taken together, these data indicate that ELS negatively impacts both males and females, but for fear discrimination this effect is not uniform across the sexes.

Disclosures: R.A. Zacharias: None. **C. Andreansky:** None. **M.H. Ray:** None. **M.A. McDannald:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.08/RR6

Topic: G.01. Appetitive and Aversive Learning

Support: ImPact

Title: An investigation into the long term and cross- contextual effects of fear counter-conditioning via DecNef

Authors: *J. E. STEWART[1], M. KAWATO[2], A. KOIZUMI[3], H. LAU[4]
[1]Computat. Neurosci., ATR, Nara-Shi, Japan; [2]Computat. Neurosci., ATR, Kyoto, Japan; [3]CiNet, NICT, Suita, Japan; [4]Univ. of California, California, CA

Abstract: Acquired fear responses to simple visual stimuli have been successfully reduced by a novel counter-conditioning procedure where decoded neurofeedback (DecNef) was used to pair reward with the multi-voxel neural patterns that represent these stimuli (Koizumi et al., 2016). This procedure could potentially benefit patients with fear-related disorders who cannot tolerate the direct encounters with fear-associated stimuli that occur during conventional fear reduction procedures. However, clinical application of this type of DecNef awaits further investigation to clarify whether the fear reduction effect (1) can be achieved with more naturalistic stimuli, (2) survives changes in context, and (3) is maintained over time. We have therefore begun a new series of experiments where we use angry faces as naturalistic visual stimuli , where we use different odors to manipulate context, and where we re-test fear responses three months after original testing. We aim to examine whether acquired fear responses to angry face stimuli can be

reduced via DecNef counter-conditioning, and whether these potential fear reduction effects are maintained across different odor contexts and over time. The results of preliminary experiments, as well as plans for future experiments, will be discussed.

Disclosures: J.E. Stewart: None. **M. Kawato:** None. **A. Koizumi:** None. **H. Lau:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.09/RR7

Topic: G.01. Appetitive and Aversive Learning

Support: Swiss National Science Foundation

Title: Modelling the impact of matrix metalloproteinase inhibition on human threat learning

Authors: *F. MELINSCAK[1], A. TZOVARA[1,2], D. R. BACH[1,2]
[1]Div. of Clin. Psychiatry Res., Psychiatric Hospital, Univ. of Zurich, Zurich, Switzerland; [2]Wellcome Trust Ctr. for Neuroimaging, London, United Kingdom

Abstract: Long-term potentiation is a fundamental memory mechanism and requires a signalling cascade that involves matrix metalloproteinase (MMP) 9. We have previously shown that the blood-brain barrier crossing, in vivo and in vitro human MMP inhibitor doxycycline impairs human threat learning (fear conditioning), as assessed by fear-potentiated startle in a retention test 7 days after learning [Bach, Tzovara, Vunder, 2017: Molecular Psychiatry]. Doxycycline also had an impact on skin conductance responses during acquisition, but this impact is not understood quantitatively. Here we harness computational models of aversive learning to derive a formal description of the observed learning trajectory.
Our models are designed to capture activity of the sympathetic nervous system (SNS) upon CS presentation, which is not directly observable. To obtain trial-by-trial estimates of SNS activity, we apply a psychophysiological modelling (PsPM) approach to skin conductance data. We then fit a number of computational associative learning models. Our model spaces encompasses descriptive models from reinforcement learning theory, normative Bayesian learning, and null models. Relative evidence for the models is assessed through Bayesian model selection.
Across all subjects, we find that the learning trajectory of SNS activity is best approximated by Bayesian learning with a beta-binomial model that fully embodies the task structure. The mapping from model onto SNS activity appears to reflect a mixture of the prior mean and entropy, thus replicating our previous work. The same computational model is also the most plausible one in both the placebo and doxycycline group, when analyzing the groups separately. We then proceed to identify variants of this model that may capture the impact of MMP inhibition during learning.

To summarise, we use computational models to link MMP inhibition to behavioural indices of threat learning, in order to capture trial-by-trial dynamics of aversive learning under conditions of impaired long-term potentiation.

Disclosures: F. Melinscak: None. **A. Tzovara:** None. **D.R. Bach:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.10/RR8

Topic: G.01. Appetitive and Aversive Learning

Support: R01-MH087755 to SSN

Title: Gamma oscillations in BLA - a computational perspective

Authors: *F. FENG[1], D. B. HEADLEY[2], Z. CHEN[1], B. LATIMER[1], A. AMIR[2], D. PARE[2], S. S. NAIR[1]
[1]Electrical and Computer Engin., Univ. of Missouri-Columbia, Columbia, MO; [2]Ctr. for Mol. and Behavioral Neurosci., Rutgers Univ., Newark, NJ

Abstract: The basolateral amygdala (BLA) participates in the formation and expression of emotional memories. Many of the brain regions that project to the BLA exhibit oscillations in the gamma (35-100 Hz) frequency band (Headley and Pare, 2013). In addition to these extrinsic sources of gamma drive, the BLA itself is thought to be capable of generating gamma oscillations on its own. However, the physiological mechanisms that support gamma rhythmogenesis in the BLA are unknown. Similarly, the relative contributions of the intrinsic mechanisms vs. extrinsic drive remain unclear.
We used a biophysical 1000-cell computational model of the BLA to investigate the role of intrinsic mechanisms and state-dependent extrinsic drive on the generation of gamma oscillations. The network model was developed using the known physiological and anatomical properties of the BLA, specifically different single cell types and their neurophysiology, synaptic current dynamics, spatially heterogeneous spatial connectivity, short- and long-term plasticity and neuromodulatory inputs (Rainnie et al., 1993; Woodruff and Sah, 2007; Headley et al., unpublished results).
The model replicated several features of gamma oscillations seen in *in vivo* local field potential recordings from BLA (unpublished data), with a prominent peak in the simulated local field potential at 60 Hz. The firing rates of model principal cells (PNs) and interneurons (ITNs) followed a log-normal distribution with averages of 0.79±0.59 Hz and 20.21±10.4 Hz, respectively, consistent with biological data (Amir et al., 2015). Both PN and ITN inter-spike interval distributions showed exponential decay with coefficient of variation values of 0.6 for

PNs, and 1.3 for ITNs, indicating irregular spiking. Moreover, both PNs and ITNs exhibited weak synchronization to the gamma cycle (vector strength: 0.18±0.1 for PNs, and 0.35±0.04 for ITNs), in agreement with *in vivo* recordings (unpublished data). We also note that, consistent with prior studies, generation of the weak gamma in BLA required that the drive to ITNs come primarily from PNs. On-going work examines the role of underlying biophysical mechanisms and connectivity schemes in the genesis of gamma. In parallel, we are investigating how state-dependent properties of the extrinsic drive, such as its gamma entrainment, produce BLA gamma.

Disclosures: F. Feng: None. **D.B. Headley:** None. **Z. Chen:** None. **B. Latimer:** None. **A. Amir:** None. **D. Pare:** None. **S.S. Nair:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.11/RR9

Topic: G.01. Appetitive and Aversive Learning

Support: NIH grant R21MH104018

NIH grant R43MH103936

Title: Role of PSD95 and nNOS binding in the regulation of conditioned fear

Authors: *L. LI[1,2], E. T. DUSTRUDE[3], M. M. HAULCOMB[4,3], A. R. ABREU[3], S. D. FITZ[3], S. K. FLORIO[4], A. I. MOLOSH[3,2], Y. Y. LAI[5,4], A. SHEKHAR[3,2]
[1]Program In Med-Neuro, Indiana Univ. Sch. of Med., Indianapolis, IN; [2]Paul and Carole Stark Neurosciences Res. Institute, Indiana Univ. Sch. of Med., Indianapolis, IN; [3]Dept. of Psychiatry, Indiana Univ. Sch. of Med., Indianapolis, IN; [4]Anagin, Inc., Indiana Ctr. for Biomed. Innovation, Indianapolis, IN; [5]Dept. of Psychological and Brain Sciences, Indiana Univ. Bloomington, Bloomington, IN

Abstract: Stimulation of N-methyl-D- aspartic acid receptors (NMDARs) and the resulting activation of neuronal nitric oxide synthase (nNOS) are crucial for fear memory formation. While NMDAR antagonists and NOS inhibitors can disrupt fear conditioning, they also disrupt many other CNS functions. The 95 kDa postsynaptic density protein (PSD95) is a scaffolding protein that links nNOS to NMDARs, which is required for the efficient activation of nNOS. We hypothesized that uncoupling nNOS from PSD95/NMADRs through either systemic and intra-amygdala application will reduce conditioned fear, similar to NMDAR antagonists but without other motor and learning deficits. In this study, we explore this hypothesis by utilizing a compound that disrupts PSD95/nNOS binding, ZL006 for its conditioned fear-attenuating effects

and determining the cellular mechanisms mediating such effects. First, we demonstrate that i.p. injection of ZL006 and not its inactive isomer ZL007, can attenuate fear memory, as measured with auditory fear conditioning. More importantly, unlike NMDARs antagonist MK801, systemic ZL006 is devoid of effects on locomotor activity, social interaction, object recognition memory and spatial memory. We show that co-immunoprecipitation analysis of tissue homogenates taken from rats soon after fear-conditioning showed a robust increase in the amygdala PSD95/nNOS binding, which is blocked by systemic pre-administration of ZL006. Treatment of amygdala slices with ZL006 also impairs long-term potentiation (LTP), the cellular signature of synaptic plasticity in amygdala neurons. Finally, we demonstrate that direct intra-amygdala infusion of ZL006 also attenuate fear conditioning. These findings support the hypothesis that disrupting the PSD95/nNOS interaction downstream of NMDARs reduces fear memory and may represent a novel treatment approach for fear-related disorders, such as post-traumatic stress disorder.

Disclosures: L. Li: None. **E.T. Dustrude:** None. **M.M. Haulcomb:** None. **A.R. Abreu:** None. **S.D. Fitz:** None. **S.K. Florio:** None. **A.I. Molosh:** None. **Y.Y. Lai:** None. **A. Shekhar:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.12/RR10

Topic: G.01. Appetitive and Aversive Learning

Support: NIH Grant DA034010

NIH Grant MH113053

Title: Lateral orbitofrontal cortex regulation of aversive prediction errors

Authors: *M. H. RAY, E. HANLON, M. A. MCDANNALD
Psychology, Boston Col., Chestnut Hill, MA

Abstract: The capacity to correctly discriminate cues signaling danger and safety is crucial and dysfunction of this capacity is a hallmark of anxiety disorders, including PTSD. In real life, cues do not signal danger or safety with complete certainty. Instead, uncertainty is inherent. Furthermore, uncertainty itself is not typically *static*, but is subject to change over time. Making and updating accurate estimates of threat in uncertainty is therefore essential. Here we tested the hypothesis that the lateral orbitofrontal cortex (lOFC) is necessary for employing aversive prediction errors related to changes in uncertainty. Male, Long Evans rats received bilateral neurotoxic lateral orbitofrontal cortex lesions using NMDA. Shams received identical surgical

treatment but did not receive NMDA infusions. Following recovery, rats received fear discrimination in which three different auditory cues were associated with different probabilities of foot shock. Fear to each cue was measured by nose poke suppression. The safety cue never predicted foot shock (p=0.00) while the danger cue always predicted foot shock (p=1.00). For the first 16 sessions, the uncertainty cue predicted would occur on 25% of trials (p=0.25). Rats were then subjected to shifts in the probability of the uncertainty cue in two blocks, both consisting of four sessions each. The first block was designed to induce a large, positive prediction error (Large, +PE block) by shifting the probability from 0.25 to 0.50. The second block was designed to induce a large, negative prediction error (Large, -PE block) by shifting the probability from 0.50 to 0.13.
We found that both Sham and lOFC-lesioned rats acquired the initial discrimination. In the Large, +PE the lOFC-lesioned rats generalized their increase in fear to the danger and safety cues. However, the sham rats did not generalize their increase in fear. In the Large, -PE block both Sham and lOFC-lesioned rats decreased fear to the uncertainty cue. Although now, in Sham rats the decrease was generalized to the danger cue, whereas in the lOFC-lesioned rats no generalization was apparent.
These results suggest that the lateral orbitofrontal cortex is involved in regulating the generalization of positive and negative aversive prediction errors. Being able to accurately and rapidly employ aversive prediction errors is an important part of associative learning. Incorrectly generalizing aversive prediction errors is maladaptive and may be indicative of human anxiety disorders, such as PTSD.

Disclosures: M.H. Ray: None. **E. Hanlon:** None. **M.A. McDannald:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.13/RR11

Topic: G.01. Appetitive and Aversive Learning

Support: Swiss National Science Foundation SNF 161077

NIMH grant MH105515

Klingenstein-Simons Fellowship Award in the Neurosciences

Title: Attenuated threat reversal learning in combat veterans

Authors: *P. HOMAN[1], E. FELTHAM[2], J. HU[1], J. H. KRYSTAL[2], I. LEVY[3], I. HARPAZ-ROTEM[2], D. SCHILLER[4]
[1]Dept. of Psychiatry, Icahn Sch. of Med. At Mount Sinai, New York, NY; [2]Dept. of Psychiatry,

Yale Univ. Sch. of Med., New Haven, CT; [3]Section of Comparative Med., Yale Sch. of Med., New Haven, CT; [4]Psychiatry and Neurosci. Departments, and Friedman Brain Inst., Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Rapid assessment of threat and safety predicting cues is crucial in an ever-changing environment. When safe cues become threatening and threatening cues become safe, flexible reversal of the learned defensive responses is required. Patients with posttraumatic stress disorder (PTSD) generally show higher defensive responses to safe cues, but the crucial ability to update threat learning has not been investigated and may represent an independent objective marker of the disease. Here, we measured skin conductance response and functional resonance magnetic imaging during threat acquisition and reversal in combat veterans with PTSD (VPTSD; N=30), veteran combat controls (VCC; N=30) and healthy human controls (HCC; N=33). We hypothesized that VPTSD would show attenuated updating of learned defensive responses compared to VCC and HCC. Indeed, the reversal learning index (defined as the difference between threat acquisition and threat reversal) was attenuated in VPTSD ($t(61) = -2.10$, $P = 0.04$; Cohen's $d = -0.53$) but also in VCC ($t(61) = -2.15$, $P = 0.04$; Cohen's $d = -0.55$) compared to HCC while there was no statistical difference between veteran groups. In line with previous PTSD imaging meta-analyses we focused our neural analysis on brain regions that have shown hyperactivity (amygdala, insula) and hypoactivity (ventromedial prefrontal cortex, dorsal striatum, dorsal anterior cingulate cortex, thalamus) and found lower neural differentiation between the threatening and safe stimuli during both acquisition and reversal in the combined veteran group compared to HCC in the bilateral caudate nuclei (left: $t(79) = -2.61$, $P = 0.01$, Cohen's $d = -0.59$; right: $t(79) = -2.89$, $P = 0.005$, Cohen's $d = -0.65$) and right thalamus ($t(79) = -2.52$, $P = 0.01$, Cohen's $d = -0.57$). Differential neural activity during threat acquisition and reversal in the dorsal anterior cingulate cortex (dACC) predicted the strength of the reversal learning index in the combined veteran group ($r(48) = 0.3$, $P = 0.03$). In conclusion, the reversal index reduction in VPTSD and VCC scaled negatively with differential neural activity in the dACC, suggesting a potentially protective role of the dACC in restoring learned threat updating.

Disclosures: P. Homan: None. **E. Feltham:** None. **J. Hu:** None. **J.H. Krystal:** None. **I. Levy:** None. **I. Harpaz-Rotem:** None. **D. Schiller:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.14/RR12

Topic: G.01. Appetitive and Aversive Learning

Support: BRAIN MH109104

Title: Mapping the cholinergic engram in fear & anxiety

Authors: *P. M. RAJEBHOSALE[1,2], D. TALMAGE[1,3], L. W. ROLE[1,4]
[1]Ctr. for Nervous Syst. Disorders, State Univ. of New York At Stony Brook, Stony Brook, NY; [2]Program in Neurosci., [3]Dept. of Pharmacol. Sci., [4]Dept. of Neurobio. & Behavior, Stony Brook Univ., Stony Brook, NY

Abstract: Anxiety disorders are one of the most common psychiatric diagnoses around the globe and are often comorbid with a variety of other mental illnesses (Kessler et al. 2005). Numerous studies have pointed to smoking cigarettes and nicotine dependence as risk factors for developing anxiety disorders (Moylan et al. 2012); some of the most illuminating studies are those of nicotine dependence in subjects with PTSD (Beckam et al. 1995. Koenen et al. 2005). In the brain, nicotine binds the nicotinic acetylcholine receptor (nAChR), the endogenous ligand for which is acetylcholine (ACh). Additionally, the basolateral amygdala (BLA), which plays an important role in fear learning, receives dense cholinergic innervation from basal forebrain cholinergic neurons (BFCNs). In the process of investigating the role of cholinergic signaling in fear learning and memory, our lab has recently demonstrated that acetylcholine release in the basolateral amygdala (BLA) can contribute to the fidelity of fear memories after an auditory fear conditioning task (Jiang et al. 2016). To further understand the contribution of the BFCN to fear learning and recall, we have developed novel tools for activity-dependent (AD), cell-type specific (Cre dependent), and temporally controlled labeling of neurons (Hereon referred to as ADCD). Here we demonstrate the validity of using these genetic constructs for mapping cell bodies as well as terminal fields of BFCNs activated during fear learning and recall of the fear memory. Upon recall of the fear memory, we discovered BFCNs in the nucleus basalis and substantia innominata (nbM/SI) labeled with the ADCD. This region of the basal forebrain sends dense projections to the BLA. Preliminarily, we observe variations in learning and/or memory between individual animals, which correlate with the number of ADCD positive BFCNs. These data suggest that there might be a correlation between cholinergic activity in the nBM/SI and either the ability of the animal to learn and/or recall the fear memory. *Prima facie*, there also seems to be an anterior-posterior bias to where these activated cholinergic neurons are found within the basal forebrain.

Disclosures: P.M. Rajebhosale: None. **D. Talmage:** None. **L.W. Role:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.15/RR13

Topic: G.01. Appetitive and Aversive Learning

Title: Cued fear memory discrimination and generalization over time

Authors: *J. BEZEK, G. POLLACK, L. WEINGAST, H. BERGSTROM
Psychological Sci., Vassar Col., Poughkeepsie, NY

Abstract: The ability to properly generalize learned stimuli is adaptive, whereas over-generalization is thought to contribute to anxiety disorders and post-traumatic stress disorder. The passage of time is one factor that can influence memory generalization. Fear conditioned contextual stimuli are well-known to generalize over time. Surprisingly little is known about the extent of generalization for auditory fear conditioned stimuli with time. The present study sought to establish an auditory fear conditioned stimulus discrimination gradient at 1-day (recent) and 30-days (remote) following learning. Adult male C57BL/6 mice were fear conditioned with three auditory conditioned stimuli (CS; 75 dB, 5 kHz, 20 s) that each co-terminated with a foot shock unconditioned stimulus (US; 0.5 s, 0.6 mA) in context A. One day after fear conditioning, mice were presented with one of five tones at different frequencies (2, 3, 5, 8, or 12 kHz; 75 dB, 20s) in a novel context (context B). 30 days later, mice were returned to context B and replayed the tone. Results showed mice discriminated most (2, 3 and 12 kHz), but not all (8 kHz), auditory frequencies at the recent frame (24 hrs). Mice were still able to discriminate the same stimuli 30 days later, suggesting a high degree of accuracy for cued fear memory retrieval over time. In a second experiment, the tone (2, 3, 8, 5 and 12 kHz) was played at only the remote time point following learning. In contrast to the first experiment, results showed a flattening of the stimulus generalization gradient, with significant cued-specific generalized responsivity occurring at the 3 kHz frequency. Based on these results, we conclude that the sharpness of the remote auditory cued fear memory generalization gradient depends on pre-exposure to the non-target stimuli at a recent time point following learning. Measurements of Arc/arg 3.1 neuron density at recent and remote time points in the lateral amygdala, temporal association cortex and prelimbic cortex is ongoing.

Disclosures: J. Bezek: None. **G. Pollack:** None. **L. Weingast:** None. **H. Bergstrom:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.16/RR14

Topic: G.01. Appetitive and Aversive Learning

Support: NIH Grant M0H38774.

Title: Mediated generalization in active avoidance

Authors: *V. CAMPESE[1], T. J. MARTIN[1], C. H. ROBERTS[1], C. DRAUS[1], J. E. LEDOUX[1,2]
[1]Ctr. For Neural Sci., New York Univ., New York, NY; [2]Emotional Brain Inst., Nathan Kline Inst. for Psychiatric Res., Orangeburg, NY

Abstract: In Pavlovian to instrumental transfer (PIT) a conditioned stimulus (CS) that has been paired with an unconditioned stimulus (US) energizes goal directed behavior. PIT has typically been studied using appetitive reinforcement, and has provided an effective model that has advanced our understanding of the psychobiological substrates of appetitive motivation. However, we still know relatively little about aversive motivation. Aversive versions of PIT typically employ unsignaled active avoidance behavior and demonstrate control over this class of responding by a CS for an aversive US (e.g., shock). The psychological and neural mechanisms underlying aversive PIT are not well understood but early findings indicate it is a unique form of avoidance behavior. In order to further explore aversive PIT we evaluated whether generalization known as acquired equivalence is involved in transfer. In this framework, stimuli that predict the same US come to be treated as similar, increasing generalization between them. To explore if context-shock and tone-shock pairings contribute to this, subjects were exposed to two avoidance chambers during training. In context A (a standard avoidance chamber) shuttling was acquired as a footshock avoidance response using an unsignaled Sidman procedure, whereas in context B (another avoidance chamber with checkered walls and smooth floors), shock was never delivered. A shock-paired CS produced less transfer (i.e., PIT) in context B than in context A. This suggests that at least in part, the aversive PIT effect is driven by the avoidance context and may be due to generalization via shock. We followed this by testing if PIT requires regions of the brain that have been implicated in spatial learning (e.g., hippocampus) and mediated generalization (e.g., entorhinal cortex). Designer hM4Di receptors were used to inhibit these regions during PIT testing, but found no behavioral effect. Subjects then underwent training on a negative patterning task, which studies have found involve both hippocampal and entorhinal regions. Inhibition of these regions via treatment with CNO (the ligand for designer receptors) impaired acquisition of the negative patterning discrimination relative to vehicle controls and a non-viral CNO control group. This pattern of findings suggests that these regions may be involved in learning based on special forms of generalization, but since aversive PIT is a performance phenomenon, these regions are not required.

Disclosures: V. Campese: None. **T.J. Martin:** None. **C.H. Roberts:** None. **C. Draus:** None. **J.E. LeDoux:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.17/RR15

Topic: H.01. Animal Cognition and Behavior

Title: Retention intervals enhance associative competition produced by a preexposed conditioned stimulus

Authors: *T. SCHACHTMAN[1], D. KLAKOTSKAIA[2], R. A. RICHARDSON[2], A. TAMASI[2], C. BAKER[2]
[2]Psychological Sci., [1]Univ. of Missouri, Columbia, MO

Abstract: Earlier studies have shown that a latent inhibitor is poor at competing for learning with another conditioned stimulus (CS) on a compound conditioning trial. Previous research also has shown that the poor conditioned response (CR) produced to a latent inhibitor can be reversed by a retention interval placed after conditioning and prior to testing the CR (e.g., Bakner et al., 1991). In the present conditioned taste aversion experiments, a CS ("A") was given CS-alone preexposures prior to a pairing of the CS with the unconditioned stimulus as pretraining phases. A compound conditioning phase also occurred in which this CS was able to compete with an added novel CS ("B"). However, prior to the AB-US compound conditioning phase, a retention interval occurred lasting either one day or many days (15 or 21 days). It was found that the lengthy retention interval enhanced the competitive potential of the pretrained CS. These results show that treatments that enhance the expression of a CS-US association can also enhance the competitive ability of the CS. However, a confirmation of the Bakner et al. effect in which the retention interval enhances the CR has not yet been examined in our laboratory using the present procedure.

Disclosures: T. Schachtman: None. **D. Klakotskaia:** None. **R.A. Richardson:** None. **A. Tamasi:** None. **C. Baker:** None.

Poster

417. Fear and Aversive Learning and Memory: Modulation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 417.18/RR16

Topic: G.01. Appetitive and Aversive Learning

Support: NIMH grant MH105515

Klingenstein-Simons Fellowship Award in the Neurosciences

Swiss National Science Foundation SNF 161077

Title: Affective flexibility without perceptual awareness

Authors: *D. SCHILLER[1], P. HOMAN[3], H. LAU[2], C. M. RAIO[4], D. R. BACH[5], D. CARMEL[6]
[1]Psychiatry and Neurosci. Departments, and Friedman Brain Inst., [2]Icahn Sch. of Med. at Mount Sinai, New York, NY; [3]Dept. of Psychiatry, Icahn Sch. of Med. At Mount Sinai, New York, NY; [4]New York Univ., New York, NY; [5]Univ. of Zurich, Zurich, Switzerland; [6]Univ. of Edinburgh, Edinburgh, United Kingdom

Abstract: In an ever-changing environment, cues that predict threat constantly change – new cues become threatening while old ones become safe. Affective flexibility is the ability to dynamically update our affective responses when threat contingencies change. Yet, confronting threatening cues in order to modify our responses could be difficult. Is it possible to achieve affective flexibility without consciously perceiving the threatening cues? We addressed this question using threat reversal learning with or without perceptual awareness of the conditioned cues, in 101 healthy human participants. To prevent perceptual awareness, we used continuous flash suppression, a technique allowing suppression of stimuli from awareness for long durations (seconds to minutes). Both aware and unaware participants were able to accomplish a dynamic updating process by shifting learned defensive responses from a stimulus that no longer predicts threat to one that does. Baseline anxiety levels scaled negatively with the strength of this updating in unaware participants. A trial-by-trial analysis of individual awareness reports, however, revealed in some participants patterns that appear unlikely under complete stimulus suppression, which conventional analyses for assessing perceptual awareness used to date might easily miss. Nevertheless, our results survived exclusion of the corresponding participants. We conclude that measures of perceptual awareness ought to consider individual trial-wise patterns, and that the successful updating of learned defensive responses requires affective flexibility but not perceptual awareness.

Disclosures: D. Schiller: None. **P. Homan:** None. **H. Lau:** None. **C.M. Raio:** None. **D.R. Bach:** None. **D. Carmel:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.01/RR17

Topic: G.02. Motivation

Support: MH107648

MH093672

Title: Dopamine D2 receptor upregulation in cholinergic interneurons (CINs) of the nucleus accumbens: Effects on CIN function and Pavlovian incentive motivation

Authors: *E. F. GALLO[1], *E. F. GALLO[1], E. TEBOUL[2], M. BAILEY[3], P. BALSAM[4], J. A. JAVITCH[3], C. KELLENDONK[2]
[1]Psychiatry, [2]Columbia University/NYSPI, New York, NY; [3]Columbia Univ., New York, NY; [4]Columbia University/Barnard Col., New York, NY

Abstract: Alterations in striatal dopamine D2 receptor (D2R) availability are associated with several neuropsychiatric disorders that feature motivational dysfunction. Cholinergic interneurons (CINs), which express D2Rs, account for 2-3% of the striatal cell population, but exert widespread control over striatal circuit function. *In vivo*, CINs exhibit a dopamine-dependent pause in firing activity in response to reward-related stimuli that is deemed critical for cue-reward associations. *In vitro* studies have implicated the D2R in mediating the dopamine-dependent pause recorded in CINs. However, conventional pharmacological tools are limited in their ability to disentangle the specific contribution of CIN D2Rs to the pause and to incentive motivation given that other cell types that operate within striatum to process reward also express D2Rs. Here, we used a Cre-dependent viral strategy to selectively express either D2Rs or EGFP in CINs of the mouse nucleus accumbens (NAc). A general Pavlovian-to-Instrumental transfer (PIT) paradigm was used to measure the motivational influence of reward-associated cues on instrumental responding for food. Our preliminary data show that, during PIT testing, responding during pre-CS+ and the CS+ periods was comparable between D2R- and EGFP-expressing mice. However, D2R upregulation led to reduced overall responding to the CS- compared to controls. This effect was particularly significant in the first of four CS+/CS- presentations. These data suggest that, rather than invigorate responding during a reward-related cue, D2R upregulation in CINs of the NAc may enhance the inhibition of actions co-occurring with a neutral cue. Because reward-related cues in PIT elicit increases in phasic dopamine release in the NAc, we sought to determine whether increased D2R expression in CINs alters their response to dopamine release. To this end, we used a similar approach to express D2Rs or EGFP in CINs of the NAc while expressing ChR2 in midbrain dopamine neurons. Using acute slices, we measured the duration of the pause in CIN firing evoked by 20 Hz photostimulation of dopamine afferents. CINs overexpressing D2Rs showed a significant pause elongation compared to EGFP-expressing CINs (D2R: 2.0 ± 0.30 s; EGFP: 0.76 ± 0.15 s; $p < 0.005$), without altering the average interspike interval. Blocking D2Rs with sulpiride abolished the pause in both conditions, indicating that D2Rs are necessary for the light-evoked pause in CINs. Together, these results suggest that CIN-selective D2R upregulation is associated with a prolonged pause response to burst firing of dopamine neurons, which may underlie the effect on Pavlovian incentive motivation.

Disclosures: E.F. Gallo: None. **E. Teboul:** None. **M. Bailey:** None. **P. Balsam:** None. **J.A. Javitch:** None. **C. Kellendonk:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.02/RR18

Topic: G.02. Motivation

Support: NIH DA016285-4

Title: Effects of environmental enrichment on sucrose cue reactivity and ventral striatal and cortical DARPP32 and DARPP32Thr34 in rats

Authors: *J. W. GRIMM, E. GLUECK, D. GINDER, J. HYDE, K. NORTH, H. REISTERER, J. SULC, K. JIGANTI
Dept of Psych and Beh Neurosci, Western Washington Univ., Bellingham, WA

Abstract: Background: Acute (17h) or chronic (30d) environmental enrichment (EE) reduces sucrose taking and seeking in rats. Our study examined the effects of acute or chronic EE on sucrose seeking and brain region levels of dopamine- and cAMP-regulated neuronal phosphoprotein molecular mass 32 kDa (DARPP32) and DARPP32 phosphorylated at threonine 34 (DARPP32Thr34). Changes in DARPP32 regulation may reflect altered dopamine (DA) D1 (DAD1) receptor-mediated signaling. Methods: Male Long-Evans rats first underwent 10 days (2h/day) of sucrose self-administration training. Control (CON) subjects remained in standard, single housing for the remainder of the experiment. Other rats experienced overnight (acute) or chronic (30d) EE (ChronicEE). Acute EE was provided immediately after the 10^{th} day of training (Day 1 AcuteEE) or 29 days later (Day 30 AcuteEE). Some rats responded for sucrose cues for 1h (TEST) immediately prior to sacrifice and brain extraction while others were sacrificed at a similar time without testing (NO TEST). Micropunches from several mesocorticolimbic regions were run through the western blot assay to identify intensity of signal for antibody-labelled DARPP32 and DARPP32Thr34. Reported here are results from ventral striatum (nucleus accumbens core and shell) and cortex (anterior cingulate and infralimbic cortices). Results: Regardless of TEST or NO TEST condition, in the nucleus accumbens core DARPP32 signal intensity was reduced by either acute or chronic EE. Acute EE increased the ratio of phosphorylated to total DARPP32 intensity. In the nucleus accumbens shell, DARPP32 signal intensity was reduced only in the NO TEST Day1 AcuteEE condition. There were no significant findings in the anterior cingulate cortex. Regardless of TEST or NO TEST condition, in the infralimbic cortex DARPP32 and the ratio of phosphorylated to total DARPP32 signal intensity were highest in ChronicEE rats. For the nucleus accumbens core, Pearson's r correlations were observed between responding for a sucrose-paired cue and DARPP32Thr34 ($r = -0.36$) and the ratio of the phosphorylated to total protein ($r = -0.44$). All results were statistically significant at $P < 0.05$. Discussion: These results support a role for DAD1 receptor signaling in the anti-craving effects of EE and indicate that targets for further investigation regarding the neurobiology of the anti-craving effects of EE include DARPP32 and DARPP32Thr34 in the nucleus accumbens core and infralimbic cortex. The accumbens core may be especially important due to the correlations between the objective measure of craving and protein signal intensities.

Disclosures: J.W. Grimm: None. **E. Glueck:** None. **D. Ginder:** None. **J. Hyde:** None. **K. North:** None. **H. Reisterer:** None. **J. Sulc:** None. **K. Jiganti:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.03/RR19

Topic: G.02. Motivation

Support: Provost Undergraduate Research Award (Johns Hopkins University Undergraduate Award)

Title: Discovery of dopaminergic circuitry controlling *Drosophila* sleep and other behaviors

Authors: *S. LUU, M. C. W. HO, T. XIE, M. WU
Johns Hopkins Univ., Baltimore, MD

Abstract: Dopamine (DA) is a conserved neurotransmitter that plays crucial roles in regulating neural circuit function and behaviors in both health and disease.[1,2] This includes behaviors such as sleep/wake, arousal, and learning and memory. Genetic tools newly developed by our lab allow for intersectional strategies to target and examine the functions of small subsets of DA neurons on animal behavior.
In our studies, DA neurons are thermogenetically activated using dTRPA1 or silenced with temperature-sensitive Shibire and are assayed for effects on 1) sleep behavior using high-throughput behavioral assays and 2) gustatory learning. We have confirmed that silencing neurons within TH-D8 GAL4 driver with UAS-Shibire^{ts} results in less sleep compared to *iso31*, UAS-Shibire^{ts} control flies, which indicates that some DA neurons could promote sleep, contrary to their role in promoting wakefuless. We are using intersectional techniques to identify the specific neurons within TH-D8 GAL4 that are responsible for this phenotype. We also show that when TH-C1 GAL4 neurons are activated using UAS-dTRPA1, flies showed increased proboscis extension after training compared to *iso31*, UAS-dTRPA1 control flies, suggesting that their learning is impaired. We are using additional tools to identify the specific neurons within TH-C1 GAL4 that affect this type of gustatory learning behavior. Finally, we are also performing an unbiased video screen of various *Drosophila* behaviors using the activation of sparse DA neurons with dTRPA1. Our studies make use of novel genetic tools to dissect the DA circuitry of several fly behaviors including sleep and learning.
1. Yamamoto, K. Vernier, P. 2011. The evolution of dopamine systems in chordates. Frontiers in Neuroanatomy, 5(2), 1-21.
2. Vieillegmaringe, J. Duris, P., Geffard, M., Le Moal, M., Delaage, M., Bensch, C., Giradie, J. 1984. Immunohistochemical localization of dopamine in the brain of the insect Locusta migratoria in comparison with the catecholamine distribution determined by the histofluoresence technique. Cell and Tissue Research, 237(2), 391-400.

Disclosures: S. Luu: None. **M.C.W. Ho:** None. **T. Xie:** None. **M. Wu:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.04/RR20

Topic: G.02. Motivation

Title: Influence of repeated intracranial self-stimulation on brain-wide functional networks

Authors: *T. C. WEIDNER[1,2], D. VINCENZ[1], M. J. BROCKA[1], A. OELSCHLEGEL[1], J. GOLDSCHMIDT[1,2,3], F. W. OHL[1,2,3], M. T. LIPPERT[1,2]
[1]Systems Physiol. of Learning, Leibniz Inst. for Neurobio., Magdeburg, Germany; [2]Otto-von-Guericke-University, Ctr. for Behavioral Brain Sci. (CBBS), Magdeburg, Germany; [3]Otto-von-Guericke-University, Inst. of Biol., Magdeburg, Germany

Abstract: Reward learning is a dynamic process supported by the mesolimbic dopamine neurons, which are often encoding reward prediction error. Intracranial self-stimulation allows for associating an operant behavior with the stimulation of these neurons. The technique has been used widely to study the neural basis of learning, addiction and other brain functions. Over the course of the acquisition of self-stimulation behavior during training, the pressing rates increase in a fashion similar to a learning curve. However, the process can also be seen in terms of a developing addiction-like condition. Here, we investigate the brain-wide plastic changes in functional network activity, which accompany this process. We transduced TH::Cre- and DAT::Cre-positive mice with ChR2(H134R) and implanted a fiber into the left VTA. The animals were then trained to perform an intracranial self-stimulation task over the course of two weeks. We subsequently performed regional single-photon emission computed tomography imaging of regional cerebral blood flow (^{99m}Tc-HMPAO SPECT) to investigate the resting state network activity in the absence of optogenetic stimulation. In the control group, mice were implanted with fibers and optogenetically transduced, but did not undergo self-stimulation training nor were they ever stimulated. Both groups showed marked differences in resting state activity levels due to the experience of self-stimulation training. In particular, TH::Cre mice exhibited elevated levels of nucleus accumbens activity in the trained group. In contrast, DAT::Cre mice showed the opposite effect: a suppression of nucleus accumbens activity after training. In general, our findings demonstrate the strong, plasticity-inducing properties of VTA self-stimulation. These effects are particularly pronounced in structures linked to addiction, like nucleus accumbens and prefrontal cortex. Furthermore, the differing cellular composition of the stimulated population in Th::Cre vs. Dat::Cre mice has a strong influence on the resulting plastic changes.

Disclosures: T.C. Weidner: None. **D. Vincenz:** None. **M.J. Brocka:** None. **A. Oelschlegel:** None. **J. Goldschmidt:** None. **F.W. Ohl:** None. **M.T. Lippert:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.05/RR21

Topic: G.02. Motivation

Support: Polish National Science Centre grant PRELUDIUM 2014/15/N/NZ4/00761

Title: The role of glutamate receptor-dependent signaling in the dopamine system in reinforcement learning

Authors: *P. E. CIESLAK, J. RODRIGUEZ PARKITNA
Inst. of Pharmacol. PAS, Krakow, Poland

Abstract: Midbrain dopamine (DA) neurons, together with the major target of their projections - dopaminoceptive neurons in the frontal cortex and basal ganglia, provide a neural substrate for reinforcement learning and are involved in decision-making and action selection. Activity and plasticity in the DA system is largely dependent on excitatory glutamatergic transmission. Here, we sought to determine the role of glutamate receptors in the DA system by using genetically modified mice with cell-type specific ablation of NMDA or mGluR5 receptors in DA neurons and neurons expressing dopamine D1 receptors. Animals were tested in an adaptive decision-making task, that resembles a 'two-armed bandit problem', in which mouse is required to estimate by trial-and-error expected value of two alternatives associated with different reward probabilities (80% vs 20%). During each session reward probabilities were reversed after 60 trials. In order to maximize the long-term sum of rewards, a mouse had to select alternative with higher success probability and adapt their choices to changes in reward contingencies. We observed that disruption of NMDA receptor-dependent signaling in DA neurons caused an initial impairment in error-driven learning and reduced the likelihood of returning to previously rewarded alternative. Moreover, loss of mGluR5 but not NMDA receptors in D1 receptor-expressing neurons decreased reward sensitivity, and as a consequence frequency of choosing alternative with higher reward probability. Finally, loss of NMDA receptors in DA neurons and mGluR5 receptors in D1 neurons caused a delay in decision time and increased latency to collect reward. In conclusion, our results suggest that glutamate receptor-dependent signaling in the DA system is necessary for quick and optimal decision-making.

Disclosures: P.E. Cieslak: None. **J. Rodriguez Parkitna:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.06/RR22

Topic: G.02. Motivation

Title: Nucleus accumbens expression of pkiα may modulate the motivation to be physically active

Authors: *K. GRIGBSY
Biomed. Sci., Univ. of Missouri, Col. of Vet. Medi, Columbia, MO

Abstract: Despite the depth of knowledge concerning the health benefits exercise, little is known about the genetic factors underlying the motivation to be physically active. Voluntary exercise is known to be driven by the dopamine-reward system, specifically within the nucleus accumbens (NAc), and functions as a marker of positive motivation. To address this topic, our lab has created a selective breeding model in Wistar rats for high (HVR) and low (LVR) voluntary wheel-running behavior to recapitulate the genetic contributions for the motivation, and lack of motivation, to be physically active. Objective: We aimed to elucidate if genetic differences in PKA signaling within the NAc, in particular the role of PKIα (protein kinase inhibitor alpha), influences the motivation to voluntarily wheel-run in our model. Methods: Pharmacological antagonism of PKA in the NAc (analogous to the role of PKIα) of HVR and LVR rats was accomplished with H-89 and overnight wheel-running behavior was monitored. qRT-PCR was used to assess *PKIα, Homer1, ZIF268* and *Arc* mRNA levels in an *ex vivo* preparation of WT, LVR and HVR rat NAc slices following dopamine receptor type-1 (D1R) activation via DRD1 agonist, SKF-38393, and dopamine receptor type-2 activation following DRD2 agonist, Quinpirole. Western blots were used to assess CREB phosphorylation following the same DRD1 activation Results: Intra-nucleus accumbens injections of H-89 increased overnight running in HVR rats at 2.5μg/0.5μl, whereas it decreased running in LVR rats at 10μg/0.5μl. Baseline mRNA expression of *Drd1*, *PKIα,* and immediate early genes similar in WT and HVR rats, and lower in LVR rats. Both WT and HVR rats showed either no or decreased expression following Drd1 activation at 10μM and 20μM. LVR showed increased expression of most genes following Drd1 activation at 20μM. Following Drd2 activation, WT and HVR rat slices showed either no or decreased mRNA expression, whereas LVR tended to show an increase in mRNA expression at 10μM. HVR showed a marked increase in CREB phosphorylation following D1R agonism, with no increase in CREB phosphorylation in LVR slices. Significance: In light of the epidemic rise in physical inactivity worldwide, understanding the molecular and genetic differences between HVR and LVR rats, for instance the role of PKIα and other downstream targets, offers keen insight into the mechanisms underlying the motivation to be physically active.

Disclosures: K. Grigbsy: None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.07/RR23

Topic: G.02. Motivation

Support: EU 7th Framework Programme Grant 607310

Title: A novel task to study loss of control over food intake in rodents

Authors: *J. P. VERHAREN[1], M. C. M. LUIJENDIJK[1], L. J. M. J. VANDERSCHUREN[2], R. A. H. ADAN[1]
[1]Brain Ctr. Rudolf Magnus, Translational Neurosci., Utrecht Univ. Med. Ctr., Utrecht, Netherlands; [2]Utrecht University, Fac. of Vet. Med., Utrecht, Netherlands

Abstract: Loss of control over behavior is a hallmark feature of many neuropsychiatric disorders, including bulimia nervosa, addiction, and obsessive-compulsive disorder. The last decades have seen a growing research interest on this topic, and several behavioral tasks for rodents have been developed to model loss of control in human psychiatric diseases. However, these tasks are often complex, and require lengthy training in rodents. Here, we sought to overcome these drawbacks, and developed a novel behavioral task that explicitly tests the ability of a rat to inhibit its urge to consume a visually present sucrose pellet during the playback of an auditory tone. During tone trials, one can distinguish successful behavioral inhibition trials, in which the rat was able to wait with reward consumption until tone termination, and loss-of-control trials, in which the rat retrieved the reward and was therefore punished with footshock. Thus far, we have used this task to study the contributions of midbrain dopamine (DA) and the prefrontal cortex to behavioral (dis)inhibition. In view of the role of dopamine in incentive motivation and impulse control, we first measured the activity of DA neurons in this task, by performing fiber photometry in the VTA of TH::Cre rats. Surprisingly, we found no changes in DA activity during either successful or unsuccessful waiting trials, inconsistent with a role for DA in control over behavior. In line with this finding, pharmacological DA stimulation did not affect performance in the task. Second, considering the role of the prefrontal cortex in behavioral inhibition, we asked which prefrontal subregions are involved in this process. To this aim, we performed pharmacological inactivations of different prefrontal cortex regions (prelimbic, inframlimbic, lateral orbital, and medial orbital cortex) and studied its effect on task performance.In sum, we present a novel task which allows for an investigation of the neural processes behind loss of control over food intake, and show proof of concept by combining it with *in vivo* calcium imaging and neuronal manipulations. In the future, this task may be used to further unravel the complex neural circuitries underlying control over behavior.

Disclosures: **J.P. Verharen:** None. **M.C.M. Luijendijk:** None. **L.J.M.J. Vanderschuren:** None. **R.A.H. Adan:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.08/RR24

Topic: G.02. Motivation

Support: KAKEN 16K16648

KAKEN 15K01837

KAKEN 16H05135

Title: Is it possible that non-attractive male mouse could get female mind?

Authors: ***Y. N. OHNISHI**, Y. KAWAHARA, Y. H. OHNISHI, A. NISHI
Kurume Univ. Sch. of Med., Kurume, Japan

Abstract: To attract female is an eternal theme for male. We challenged to search what are criteria of female mice to select a male mouse and the neural network activity when male attracts female. Moreover, we tried to hijack the neural network for making female mice to love a non-attractive male mouse with optogenetic stimulation.At first, we have to confirm whether there are attractive and non-attractive male mice. Because we can not distinguish the difference between any healthy two mice other than their body size. For the purpose, we established the exploring behavior-based monitoring system for checking female preference to male mice. Unfortunately, this method works only one time, but we were able to detect their preference to the specific male mouse and/or avoidance behavior against the non-attractive male mouse. In this condition, we examined (1) whether their preference is common among mice having different genetic background, (2) whether female prefers to the familiar male mouse, (3) whether female prefers to father of her babies, (4) whether female mice prefer the single housing attractive male mouse or female-paired housing non-attractive male mouse, (5) whether testosterone administration changes the non-attractive male mouse into attractive one. Additionally, we measured dopamine levels in the nucleus accumbens, when female mice are encountering with the non-attractive or attractive male mouse. As we found significant difference between them, we will try to mimic the response with optogenetic stimulation and examine their effects on female preference behavior against male mice.

Disclosures: **Y.N. Ohnishi:** None. **Y. Kawahara:** None. **Y.H. Ohnishi:** None. **A. Nishi:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.09/RR25

Topic: G.02. Motivation

Support: R01DA040993

Title: Lengthening the intertrial interval increases sign tracking and dopamine release to conditioned and unconditioned stimuli

Authors: *M. R. ROESCH[1], B. LEE[2], R. N. GENTRY[4], G. B. BISSONETTE[5], R. J. HERMAN[2], J. J. MALLON[2], D. BRYDEN[6], D. J. CALU[3], G. SCHOENBAUM[7], E. COUTUREAU[8], A. R. MARCHAND[9], M. KHAMASSI[10]
[1]Univ. of Maryland at Col. Park, College Park, MD; [2]Univ. of Maryland, College Park, MD; [3]Univ. of Maryland, Baltimore, MD; [4]Psychology, Univ. of Maryland, Col. Park, College Park, MD; [5]Dept. of Psychology, Univ. of Maryland at Col. Park Dept. of Psychology, College Park, MD; [6]Univ. of Maryland Col. Park, College Park, MD; [7]Natl. Inst. on Drug Abuse Intramural Res. Program, Baltimore, MD; [8]CNRS, Bordeaux Cedex, France; [9]Univ. de Bordeaux, Bordeaux, France; [10]CNRS - Univ. Pierre Et Marie Curie, Paris, France

Abstract: Recent work showing that dopamine (DA) transmission is not necessary for all forms of learning has questioned the hypothesis that phasic dopamine corresponds to a reward prediction error signal arising from a classical Model-Free system. This hypothesis has been derived from the observation that in certain individuals known as goal trackers, DA release does not conform to the classical reward prediction error signal and that blocking DA does not impair learning during Pavlovian autoshaping. The Lesaint-Khamassi model has accounted for these observations and has provided a set of experimental predictions to further its validity. Here we test the prediction that individual variability observed during Pavlovian conditioning and associated DA release can be modulated by the length of time in between trials (intertrial interval; ITI). It has been hypothesized that increasing (resp. decreasing) the ITI allows more (resp. less) time to negatively revise the value of the food receptacle leading to increased (resp. decreased) tendencies to track predictive stimuli (i.e., sign-tracking). It is predicted that, with this constant revision of value, DA release to the US will not attenuate during learning as would be expected from classical Model-Free algorithms. To test this prediction we recorded DA release in nucleus accumbens core during a common Pavlovian autoshaping procedure (8 sec lever extension followed by reward) during sessions with either long (120 s) or short (60 s) ITIs. Consistent with model predictions, lengthening the ITI (i.e., 120 s ITI group) increased sign tracking and phasic DA release during the conditioned stimulus (CS; lever extension) and reward delivery (unconditioned stimulus; US). Importantly, phasic DA release to reward delivery was

even present after learning (10 sessions x 25 trials). During conditioning with shorter ITIs (i.e., 60 s groups), goal tracking was more prominent, and DA release to the CS was weaker, and not present at the time of reward delivery after learning. These results validate the initial computational hypotheses, opening new perspectives on the understanding of inter-individual differences in Pavlovian conditioning.

Disclosures: M.R. Roesch: None. **B. Lee:** None. **R.N. Gentry:** None. **G.B. Bissonette:** None. **R.J. Herman:** None. **J.J. Mallon:** None. **D. Bryden:** None. **D.J. Calu:** None. **G. Schoenbaum:** None. **E. Coutureau:** None. **A.R. Marchand:** None. **M. Khamassi:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.10/RR26

Topic: G.02. Motivation

Support: Conacyt 220772

Title: Differential effects of dopamine receptor activation at the nucleus accumbens and the medial preoptic area on male rat sexual behavior expression of sexually exhausted male rats

Authors: L. GUADARRAMA-BAZANTE, *G. RODRIGUEZ-MANZO
Cinvestav-Sede Sur, IPN, Ciudad DE Mexico, Mexico

Abstract: Dopamine (DA) is involved in the control of motivated behaviors such as sexual behavior. Two brain regions are implicated in the dopaminergic regulation of male rat sexual behavior: the medial preoptic area (mPOA) and the nucleus accumbens (NAcc). We have recently reported that systemic administration of the nonspecific DA receptor agonist apomorphine has different effects on copulation of sexually active animals and temporarily sexually inactive males, due to sexual satiety. Thus, apomorphine almost lacked an effect in sexually experienced rats, but had clear facilitative effects on sexual behavior expression of sexually exhausted males. To determine the role played by the NAcc and the mPOA in the mediation of apomorphine-induced effects on copulatory behavior of animals with a distinct sexual condition, different doses of apomorphine (0.6, 2.0, 6.0 µg/animal) were directly infused into the mPOA or the NAcc of either sexually experienced or sexually exhausted males (24h after satiation). Apomorphine delivered to the mPOA did not modify copulation of sexually experienced animals and slightly increased the proportion of sexually exhausted animals exhibiting sexual activity. By contrast, infusion of apomorphine into the NAcc lacked of effects in sexually active rats, but reversed sexual satiety at all doses tested. After the highest apomorphine dose tested (6.0 µg/rat), all the satiated rats achieved ejaculation and resumed copulation after ejaculation and their copulatory parameters were indistinguishable from those

exhibited by sexually experienced control male rats, suggesting a full reversal of the sexual inhibitory state. Data show that activation of dopamine receptors by apomorphine either at the mPOA or the NAcc does not modify copulatory behavior of sexually active male rats. By contrast, in sexually exhausted animals the same apomorphine doses facilitated sexual behavior expression when infused into the NAcc, but lacked of an effect when infused into the mPOA. Results suggest that activation of DA receptors at the NAcc participates in the reversal of the sexual inhibitory state that characterizes sexual satiety, while DA transmission at the mPOA is not involved in this sexual inhibition phenomenon

Disclosures: L. Guadarrama-bazante: None. **G. Rodriguez-Manzo:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.11/RR27

Topic: G.02. Motivation

Support: NIAAA IRP

Title: Coincident and causal contributions of striatal dopamine to cognitive flexibility

Authors: *A. KOCHARIAN[1], A. K. RADKE[2], D. M. LOVINGER[1], Y. MATEO[1], A. HOLMES, PhD[1]
[1]Natl. Inst. on Alcohol Abuse and Alcoholism, Rockville, MD; [2]Miami Univ., Oxford, OH

Abstract: Striatal dopamine (DA) neurons encode the difference between expected and received reward as prediction errors (RPEs) which work as teaching signals to guide adaptive choices. DA dynamics vary between different striatal regions and few studies have examined the differential contributions of DA release in the nucleus accumbens core (NAcc) and dorsolateral striatum (DLS) to the ability to flexibly update behaviors and revise associations. Here, DA transients were monitored using *in vivo* fast-scan cyclic voltammetry (FSCV) in the mouse NAcc and DLS during performance of a rewarded choice-discrimination and reversal touchscreen task. DA transients in the NAcc were significantly elevated following unexpectedly-rewarded choices during early reversal, but not other task-phases. To assess the causal contribution of DAergic inputs from the ventral tegmental area (VTA) to the NAcc in choice-flexibility, the VTA-NAcc pathway was selectively photosilenced or photostimulated during early reversal. Inhibition of DA cell terminal activity during early reversal significantly increased perseverative errors. Bidirectional effects were not observed with photostimulation at DA terminals as no changes in behavioral performance were observed in this group, suggesting that augmenting DA activity may not be sufficient to facilitate performance, possibly due to a ceiling effect in the measured DA release. Collectively, our data provide further evidence of preferential recruitment of DA

input to the NAcc when expected stimulus-reward contingencies are altered and, importantly, demonstrate a necessary causal role for this DAergic signal for choice behavior to be adapted accordingly. Research supported by NIAAA IRP.

Disclosures: A. Kocharian: None. **A.K. Radke:** None. **D.M. Lovinger:** None. **Y. Mateo:** None. **A. Holmes:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.12/RR28

Topic: G.02. Motivation

Support: Wellcome Trust 095668 and 095669

Sir Henry Wellcome Fellowship 106101

Title: Projection-specific roles of dopamine neurons in decision making

Authors: *M. CARANDINI, S. SCHRÖDER, M. WELLS, C. REDDY, K. D. HARRIS, A. LAK
Univ. Col. London, London, United Kingdom

Abstract: Reward learning and value estimation rely on the phasic activity of midbrain dopamine (DA) neurons. Recent studies suggest that subpopulations of DA neurons with different projection targets could play different roles in these behaviors. To test this hypothesis, we trained mice in a perceptual decision task, and characterized the effects of pairing reward with activation of DA neurons. We then asked how these effects depend on the projection target of the activated DA neurons.
We expressed Channelrhodopsin-2 in midbrain DA neurons of DAT-Cre mice, and implanted optical fibers to stimulate their cell bodies in ventral tegmental area (VTA) or their axon terminals in striatum or medial prefrontal cortex (mPFC). We trained these mice in a perceptual decision task rewarded with water (Burgess et al., bioRxiv, 2016). In each trial, a grating appeared on the left or right side of a monitor, and the head-fixed mouse indicated its position by turning a steering wheel. By varying stimulus contrast across trials we obtained high-quality psychometric curves. After reaching stable task performance, we paired the water reward that followed one choice option with brief optogenetic stimulation.
The effects of DA stimulation depended markedly on the location of the optical fiber. Following stimulation of DA cell bodies in VTA, mice developed a tendency towards choices paired with such stimulations. This tendency affected the psychometric bias parameter, consistent with increased value of the choice option paired with DA stimulation. Following stimulation of DA

terminals in mPFC, we observed a similar, albeit weaker, behavioral effect, which we are further testing using alternative viral methods.
We observed a different effect following stimulation of DA terminals in striatum. The effect depended on which hemisphere was stimulated: mice developed a tendency to respond as if a visual stimulus ipsilateral to the stimulated striatum was present, regardless of stimulus location. This tendency changed both the bias and a lapse parameter of the psychometric function and occurred regardless of which choices (left or right or both) were paired with the stimulation. Thus, the unilateral stimulation of striatal DA terminals could not be interpreted as a change in the value of a choice option.
Together, these results illustrate the roles of DA neurons in driving perceptual choices and indicate that DA neurons projecting to different brain structures could play different roles in decision making under perceptual uncertainty.

Disclosures: M. Carandini: None. **S. Schröder:** None. **M. Wells:** None. **C. Reddy:** None. **K.D. Harris:** None. **A. Lak:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.13/RR29

Topic: G.02. Motivation

Support: NSERC Grant RGPIN2016-06703

Title: The priming effect of rewarding brain stimulation persists with pimozide

Authors: *C. EVANGELISTA, N. MEHREZ, W. G. BRAKE, P. SHIZGAL
CSBN, Concordia Univ., Montreal, QC, Canada

Abstract: Instrumental performance is invigorated by prior delivery of free rewards. In previous studies, rats ran faster to receive rewarding electrical brain stimulation in the goal box of a runway after free, non-contingent stimulation ("priming") had been delivered in the start box. Although dopamine is strongly implicated in other forms of incentive motivation, the D2/D4 blocker, pimozide, did not eliminate the effect of priming on runway performance. In the present study, the impact of pimozide on the priming effect was reassessed using an operant chamber and systematic variation of reward cost. Male, Long-Evans rats (n = 8) were implanted with electrodes aimed bilaterally at the lateral hypothalamus and were subsequently trained to lever press for rewarding electrical brain stimulation. A single response on a setup lever triggered its retraction and the extension of a separate "reward" lever, which was armed on a fixed-ratio (FR) schedule (range: FR2-32). The FR requirement increased after every block of 15 trials. Following the completion of the FR, a 0.5-s train of 300-500 μA, 0.1-ms cathodal pulses was

delivered and the reward lever was retracted. After a 30-s inter-trial interval (ITI), the setup lever extended, starting a new trial. Either two or 10 0.5-s trains were delivered during the ITI. Vehicle or pimozide (0.1, 0.2, 0.5 mg/kg) was administered three h prior to testing. Tests were conducted in three-day cycles consisting of a vehicle day, pimozide day, and washout day. Response vigor, measured as the inverse of the latency to complete the FR requirement, decreased as the FR was increased. The curve relating response vigor to the FR was shifted leftward along the FR axis by pimozide, indicating that the drug decreased the effectiveness of the electrical reward. Nonetheless, the priming effect remained intact after drug administration: the boost in response vigor due to increasing the number of priming trains from two to 10 was similar in the drug and vehicle conditions. These results are consistent with the previous finding obtained using the runway task. Pimozide does not appear to alter the priming effect.

Disclosures: C. Evangelista: None. **N. Mehrez:** None. **W.G. Brake:** None. **P. Shizgal:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.14/RR30

Topic: G.02. Motivation

Title: Distinguishing movement from prediction signals in midbrain dopamine neurons during Pavlovian acquisition

Authors: *L. T. CODDINGTON[1], J. T. DUDMAN[2]
[1]Neurobio., HHMI Janelia Res. Campus, Ashburn, VA; [2]HHMI, Ashburn, VA

Abstract: Debates about the specifics of mammalian midbrain dopamine (mDA) reward-encoding often turn on the question of whether the signal is structured to affect ongoing behavior or to guide learning for future behavior. Recently, recordings made outside of the context of cue-reward associations suggested that mDA neurons may also represent the initiation of movement. Thus, it was proposed that mDA activity acts to control movement initiation. However, given that movement initiation signals are largely absent in well-trained animals, we note an alternative possibility: in the absence of well-represented goals, self-initiated actions might hold relatively large reward-predictive value. mDA movement initiation signals in relatively untrained animals could then reflect a transient phase in which movements generate relatively strong reward prediction signals in DA neurons. This should be evident during the transition from naivety to the acquisition of appetitive responses during Pavlovian conditioning.
We simultaneously monitored the activity of identified mDA neurons with quantitative measures of appetitive responding (body movements and licking) in head fixed mice during initial acquisition of a Pavlovian conditioned stimulus (CS; a 500 ms pure tone). We observed conditioned approach movements that together with licking served as a sensitive, continuous

learning index. We performed juxtacellular recordings from optogenetically-identified DA neurons in both the ventral tegmental area (n=48) and the substantia nigra (n=85), densely sampled throughout initial learning.
We observed phasic modulations of both SNc and VTA DA neurons to water reward delivery, reward-predictive stimuli, and self-initiated movements, consistent with disparate previous works. Responses to movement initiation were not continuously correlated with parameters of movement, but within individual cells responses to movement, CS-, and US-reward cues were correlated, reflecting a common, predictive signal subject to cell- and state-specific gain factors. CS-responses and modulation of reward encoding by expectation appeared late in acquisition learning, lagging the emergence of associated behavioral changes. Together these data support a model in which the phasic responses of mDA neurons reflect learning and are positioned to play a role in adapting appetitive behavior rather than driving associative learning per se.

Disclosures: L.T. Coddington: None. **J.T. Dudman:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.15/RR31

Topic: G.02. Motivation

Support: NIH- ZIA-DA000587

Title: Blockade of dopaminergic transients prevents learning driven by changes in reward features

Authors: *C. CHANG, M. GARDNER, M. GONZALEZ DI TILLIO, G. SCHOENBAUM
NIDA, Baltimore, MD

Abstract: Abstract:
Prediction errors are critical for associative learning. Transient changes in dopamine neuron activity correlate with positive and negative reward prediction errors and can mimic their effects. However, while causal studies show that dopamine transients are sufficient to drive learning, they do not address whether they are necessary. Further, the precise nature of this signal is not yet fully established. While it has been equated to the cached-value error signal proposed to support model-free reinforcement learning, cached-value errors are typically confounded with errors in the prediction of reward features. Here we used optogenetic and transgenic approaches to prevent transient changes in midbrain dopamine neuron activity during the critical error-signaling period of two unblocking tasks. In one, learning was unblocked by adding unexpected reward, a manipulation expected to induce errors in predicting both value and reward features. In another, learning was unblocked by switching from one to another equally valued reward, a

manipulation expected to induce errors only in reward feature prediction. Optogenetically suppressing the firing of dopamine neurons in the ventral tegmental area for 5s at the time of the changes in reward prevented unblocking of learning in both tasks. This result suggests that dopamine transients play a general role in error signaling rather than being restricted to only signaling errors in value as currently hypothesized.

Disclosures: C. Chang: None. **M. Gardner:** None. **M. Gonzalez Di Tillio:** None. **G. Schoenbaum:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.16/RR32

Topic: G.02. Motivation

Support: NSERC

Title: Can high fructose corn syrup alter behavioral and neural responses to oxycodone?

Authors: *M. MINHAS, ***M. MINHAS**, E. STROM, C. L. LIMEBEER, F. LERI
Dept. of Psychology & Collaborative Specialization in Neurosci., Univ. of Guelph, Guelph, ON, Canada

Abstract: Objective: The food addiction hypothesis suggests that refined sugars such as high fructose corn syrup (HFCS) can promote addictive-like behaviors. This leads to the prediction that sugars and drugs of abuse interact at both neural and behavioural levels. The present study examined whether rats chronically pre-exposed to high fructose corn syrup (HFCS; 55% fructose to 45% glucose) altered responses to oxycodone (OXY), a widely abused prescription opioid. Since sugars can activate opioid receptors in the ventral tegmental area to enhance dopamine (DA) release in the nucleus accumbens (NAc) to affect reward processes, we investigated whether chronic HFCS pre-exposure affected responses to: OXY- place preference, OXY-induced locomotor sensitization, and OXY-induced elevation in DA concentrations in the NAc. Methods: Male Sprague-Dawley rats received in their home cages: 0% HFCS 24h a day (n = 70); 50% HFCS 24h a day (continuous access group, CONT; n = 84); or 50% HFCS 12h a day (intermittent access beginning 4 h into the dark cycle, INT; n = 28) for 26 days. INT sugar access was employed as it leads to “bingeing” behaviour which is linked to the neural alterations that cause enhancements in reward and motor responses to stimuli. Following a 9-day sugar free period, animals were tested on one of the following: (1) place conditioning (biased design) involving: pre-test, place conditioning (0, 0.16, or 2.5 mg/kg OXY SC; 3 pairings each over 6 days), and a test of preference; (2) locomotor sensitization involving context-dependent treatment with OXY (0, 0.16, or 2.5 mg/kg SC) over 5 days, followed by a 9-day drug-free

period, following which locomotor activity was measured in the drug-paired context, both in the absence and 24 hrs later in the presence of OXY; (3) in vivo microdialysis of extracellular DA in the NAc during a 1hr baseline and 3 hrs following an injection of OXY (0 or 2.5 mg/kg SC). Results: It was found that 0.16 and 2.5 mg/kg OXY produced a place preference, but this was not modified by HFCS pre-treatment. Furthermore, HFCS pre-treatment decreased OXY-induced, but not context-induced locomotion. Microdialysis data revealed that HFCS pre-treatment decreased the OXY-induced dopaminergic response in the NAc.
Conclusion: Taken together, the current experiments indicate that chronic HFCS pre-exposure blunted the psychomotor and dopaminergic response to OXY, but did not alter OXY conditioned locomotion or reward. These results suggest that opioids and sugars interact at both behavioural and neural levels, indicating that nutrition has the potential to influence some responses to opioids, and this may be relevant to the licit and illicit use of these drugs.

Disclosures: **M. Minhas:** None. **E. Strom:** None. **C.L. Limebeer:** None. **F. Leri:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.17/RR33

Topic: G.02. Motivation

Support: FONDECYT grant N° 1150244

Title: The activation of GABA-A alpha1 receptors in the ventral tegmental area potentiates the stimulation of dopamine neurons in the antero-ventral region of the ventral tegmental area induced by lateral septum stimulation

Authors: I. M. VEGA-QUIROGA, H. E. YARUR, *K. GYSLING
Pontificia Univ. Catolica De Chile, Santiago, Chile

Abstract: The mechanisms controlling activity of dopaminergic neurons in ventral tegmental area (VTA) and the anatomical location of these neurons are relevant for the expression of motivated behavior associated to reward-related signals. Anatomical and neurochemical evidence shows that several brain regions modulate the activity of VTA dopaminergic neurons. However, there is scarce knowledge of how the lateral septum (LS) modulates VTA activity. The evidence showed that LS output is pronominally GABAergic neurons, thus we hypothesized that GABAergic projections from LS to VTA inhibit GABAergic interneurons in the VTA through GABA-A alpha 1 receptors selectivity expressed in these interneurons .We performed *in-vivo* dual-probe microdialysis in anesthetized rats to measure VTA dopamine, glutamate and GABA extracellular levels after stimulating or inhibiting LS. In addition, we combined c-Fos and tyrosine hydroxylase immunohistochemistry with to reveal which VTA dopaminergic neurons

respond to LS stimulation. In addition, we injected biotin-dextran-amine (BDA), anterograde tracer in LS to reveal the topography of LS-VTA connectivity. The presence of indiplon, selective positive GABA-A alpha 1 receptor modulator, in the VTA induced a significant increase in VTA dopamine extracellular levels induced by LS stimulation. Furthermore, we observed thata LS stimulation induced significant c-Fos expression in a subset of dopaminergic neurons in the antero-ventral region of the VTA. Consistently, intra LS BDA injection labelled fibers reaching the antero-ventral region of the VTA. Taken together our data shows that LS modulates dopaminergic activity in the antero-ventral region of the VTA by inhibiting GABAergic interneurons bearing GABA-A alpha 1 receptors.

Disclosures: I.M. Vega-Quiroga: None. **H.E. Yarur:** None. **K. Gysling:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.18/RR34

Topic: G.02. Motivation

Title: Spatio-temporally heterogeneous activity of VTA projections to dorsal striatum

Authors: *A. HAMID[1], M. J. FRANK[2], C. I. MOORE[1,2]
[1]Dept. of Neurosci., [2]Brown Inst. for Brain Sci., Brown Univ., Providence, RI

Abstract: Dopamine (DA) is closely involved in both motivation and reinforcement-driven learning. Midbrain DA activity is extensively documented to encode reward prediction errors (RPEs). This RPE message is argued to be redundantly encoded among DA cells, and uniformly broadcast to downstream targets. But, many reports provide evidence for the contrary, suggesting a specialized and heterogeneous DA circuit. First, midbrain DA cells form diverse groups based on forebrain projection-target and biophysical characteristics, and are differentially recruited in response to salient cues, reward cues or aversive cues. Second, within the striatum, DA release is under strong control of local microcircuitry (e.g glutamate, acetylcholine), and the temporal dynamics of *in vivo* DA release are heterogeneous across striatal micro (and macro) domains. A thorough examination of the functional role of region-specific DA activity is lacking, but remains essential for a deeper understanding of flexible decision making.
We performed functional imaging of VTA axons in the dorsal striatum. DAT-cre mice received midbrain infusion of GCAMP6f, and implanted with imaging cannula or GRIN lenses for optical access into the striatum. After recovery from surgery, we imaged large-scale (~2-3 mm area using 1-photon microscopy) or fine scale (using 2-photon microscopy) activity of DA terminals in the striatum during task performance. Preliminary data indicates macro domains of asynchronous DA axon activity across large regions of striatal tissue. These zones are sometimes co-active in a global manner, but were observed to be decorrelated for extended temporal epochs.

Individual DA axons imaged with 2-photon microscopy also revealed spatio-temporal heterogeneity in the activity of neighboring axons separated by a few microns. Striatal DA domains observed to have asychornous spontaneous activity also displayed behaviorally relevant functional heterogeneity, ramping to delayed rewards or phasically responding to juice delivery. Together, our results provide support for distributed, heterogeneous DA signals that may serve local computational requirements.

Disclosures: A. Hamid: None. **M.J. Frank:** None. **C.I. Moore:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.19/RR35

Topic: G.02. Motivation

Support: European Research Council, ERC Advanced Grant ERC-2012-ADG_20120314 (322541) MeSSI

Swiss National Science Fund, SNSF 310030_149985

Swiss National Science Fund, SNSF 310030B_170266

Title: Midbrain neuronal activity during cue-guided spatial learning

Authors: *Y. KREMER[1], J. FLAKOWSKI[1], C. ROHNER[1], C. LÜSCHER[1,2]
[1]Dept. Basic Neurosciences - Univ. of Geneva, Geneva, Switzerland; [2]Clin. of Neurology, Dept. of Clin. Neurosciences, Geneva Univ. Hosp., Geneva, Switzerland

Abstract: Neurons of the ventral tegmental area (VTA) have been implicated in reward processing, motivation and motor output. Dopamine neurons in particular may encode reward prediction error but also locomotor activity. Stimulating dopamine neurons induces place preference, enhances spatial working memory and modulates hippocampal place fields. However whether spatial locations per se evoke firing in dopamine neurons remains elusive. Here we ask how the mesolimbic network orchestrates neural activity to generate a successful behavioral output based on reward-predicting cues present in the environment.
We designed a spatial task for freely moving mice that allows decoupling of spatial location, cue-evoked responses and reward consumption. Mice first learned to associate a light cue (CS) with the availability of a liquid reward (US). Once this association was acquired, mice could trigger the CS-US sequence by remaining for 2 seconds in a pre-defined, unmarked location in the operant chamber. During the task we recorded single-unit activity in the midbrain (of which a fraction was optogenetically phototagged) to investigate the correlations between the individudal events of the task and the underlying firing patterns contributing to the learning and execution of

the paradigm.
Our recordings show that a large fraction of midbrain neurons encodes motor output related to movement between the different task positions or actions to collect the reward (licking). Dopamine neurons, even though they maintain reliable phasic responses to either the cue, reward delivery or both, show a large diversity in their response magnitudes even at similar stages of learning.
We then trained mice to search for new locations by changing the spatial reward contingencies. In these experiments mice quickly learned to search for new rewarded positions, but the activity of DA neurons was not altered. In a second variation, we asked the mice to perform the task in the absence of light cues, solely relying on spatial information. In these experiments, mice were still able to perform the task although no responses in DA neurons could be detected.
In summary, mice associated spatial positions with the availability of rewards, but DA neurons respond to temporally precise reward-predicting cues rather than to spatial locations predicting rewards. Additionally, even though animals show clear reward expectancy by navigating to these locations, the CS reliably triggers large bursts of action potentials indicating that the spatial location did not devaluate the reward prediction error.

Disclosures: **Y. Kremer:** None. **J. Flakowski:** None. **C. Rohner:** None. **C. Lüscher:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.20/RR36

Topic: G.02. Motivation

Support: Israel Science Foundation (ISF) grants

Adelis Foundation

Allen and Jewel Prince Center for Neurodegenerative Processes of the Brain

FP-7-CIG grant

Title: The role of the reward system in attenuating tumor progression in mice

Authors: *M. SCHILLER[1], T. L. BEN-SHAANAN[1], H. AZULAY-DEBBY[1], B. KORIN[1], N. BOSHNAK[1], J. SHAKYA[1], M. A. RAHAT[1], F. HAKIM[2], A. ROLLS[1]
[1]Technion, Haifa, Israel; [2]Rambam Hlth. Care Campus, Haifa, Israel

Abstract: Increasing immune system activity is a leading target for cancer therapy. Here, we show that the anti-tumor immune response can be enhanced by activation of the brain's reward system, a key circuitry in positive emotional states. We activated the reward system of tumor-

bearing mice (Lewis Lung Carcinoma) using chemogenetics (DREADDs), and analyzed the effects on tumor growth and anti-tumor immunity. Reward system activation resulted in a 52% ± 15.1% reduction in tumor size. This effect was mediated by a specific reduction in sympathetic activity in the bone marrow, attenuating the immunosuppressive functions of myeloid derived suppressor cells (MDSCs). Given the central role of the reward system in positive emotions, these findings introduce a physiological mechanism whereby the patient's psychological state can impact cancer progression.

Disclosures: **M. Schiller:** None. **T.L. Ben-Shaanan:** None. **H. Azulay-Debby:** None. **B. Korin:** None. **N. Boshnak:** None. **J. Shakya:** None. **M.A. Rahat:** None. **F. Hakim:** None. **A. Rolls:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.21/SS1

Topic: G.02. Motivation

Support: NARSAD Young Investigator Award

Boettcher Young Investigator Award

NSF grant IOS-1557755

R03DA038734

Title: *In vivo* electrochemical and optogenetic assessment of the nucleus accumbens in the acquisition, valuation and extinction of avoidance behavior

Authors: *K. J. PULTORAK, S. A. SCHELP, G. P. KRZYSTYNIAK, D. R. RAKOWSKI, B. BUSCH, E. OLESON
Univ. of Colorado Denver, Denver, CO

Abstract: The mesolimbic dopamine (DA) system has historically been implicated in motivational processing under appetitive contexts. Within an appetitive context, mounting evidence supports that transient accumbal DA release events increase in response to reward predictive cues, represent intrinsic reward value, and causally modify reward-seeking. Less, however, is known regarding the role the nucleus accumbens (Nac) plays in an aversive context. Here we sought to investigate the role of DA in the valuation of avoided footshock using fast-scan cyclic voltammetry (FSCV), assess the causal role of DA in valuation though optical stimulation of DA neurons, and further assess the role of DA in the acquisition and extinction of avoidance behavior utilizing optogenetic stimulation. In order to address these research

questions, we first developed a novel behavioral economics task in which rats are provided the opportunity to operantly avoid electrical footshock across epochs wherein unit-price (response requirement/mA shock avoided) increases on a semi-exponential array. In congruence with previous appetitive research, the concentration of DA observed at the avoidance-predictive cue as well as upon successful avoidance scaled in an inversely to unit-price; however, an initial suppression of dopamine release and avoidance was observed at session onset. To assess causality, we optogenetically activated ChR2 expressing DA neurons within the VTA to selectively augment DA release at either cue onset or upon successful avoidance. We observed that augmentation of VTA DA at the cue decreases the maximal price paid to avoid footshock; whereas augmenting release at successful avoidance increases the maximal price paid to avoid footshock. Next we augmented dopamine during the acquisition and extinction of avoidance either at the cue or at successful avoidance. Preliminary data suggest faster rates of acquisition, but slower rates of extinction when dopamine release is augmented at avoidance. Conversely, augmentation at the cue produce slower acquisition, but increased rates of extinction. Finally we sought to evaluate the role of other predominate input structures onto the Nac, most notably the basolateral amygdala (BLA) which has been strongly implicated in anxiety and fear learning. Preliminary data suggest that terminal augmentation of BLA glutamate neurons at the cue suppresses the acquisition of avoidance. Together, these findings suggest that transient, accumbal DA release events play an integral role in the learning, extinction and valuation of avoidance and further demonstrate that BLA-Nac neuronal circuitry influence the acquisition of avoidance.

Disclosures: K.J. Pultorak: None. **S.A. Schelp:** None. **G.P. Krzystyniak:** None. **D.R. Rakowski:** None. **B. Busch:** None. **E. Oleson:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.22/SS2

Topic: G.02. Motivation

Support: NIH Intramural Research Program (NIDDK)

Title: Food seeking engages distinct calcium responses in direct and indirect pathway striatal neurons

Authors: *T. LONDON, A. KRAVITZ
Natl. Inst. of Health/ NIDDK, Bethesda, MD

Abstract: Feeding is driven by sensory inputs related to food cues and internal hunger signals reflecting nutrient levels and energy balance. Dopamine signaling lies at the nexus of these signals, as dopamine neurons are activated by food cues and are also modulated by hunger

centers in the brain. The striatum plays a role in influencing food reinforcement through the activity of two projection neurons, direct pathway medium spiny neurons (dMSN) and indirect pathway medium spiny neurons (iMSN). These populations express different dopamine receptors, D1 and D2, respectively, and can therefore respond to dopamine differently. Based on literature implicating dMSNs in positive reinforcement and iMSNs in punishment, we hypothesized that dMSNs would be excited by cues, actions directed toward food, and food consumption, while iMSNs would be inhibited during these behavioral events. We examined the role of each neuron type in a modified Pavlovian task, while recording their activity with fiber photometry: a technique that allows for the recording of bulk calcium signals in specific cell populations. Using this technique, we quantified the responses of dMSNs and iMSNs to cues that predict food, movement to the food, and food consumption. We found food seeking behavior activated dMSNs, but not iMSNs. Counter to our hypothesis, food predictive cues did not alter population calcium signals in either pathway; while consumption strongly inhibited calcium signals in both pathways. We conclude that medium spiny neurons respond differently during the phases of food seeking and consumption. Understanding the dynamics of striatal neurons during food seeking and consumption will shed light on how the brain processes food related information, and what drives animals to seek food.

Disclosures: **T. London:** None. **A. Kravitz:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.23/SS3

Topic: G.02. Motivation

Support: NIH R01 MH080243

Title: Dynamic changes in striatal dopamine predict reward learning: Evidence from simultaneous PET/MR

Authors: *F. J. CALABRO[1], D. F. MONTEZ[2], V. P. MURTY[2], B. LARSEN[3], J. C. PRICE[4], B. LUNA[5]
[1]Departments of Psychiatry and Bioengineering, [2]Psychiatry, [3]Univ. of Pittsburgh, Pittsburgh, PA; [4]Radiology, Massachusetts Gen. Hosp., Boston, MA; [5]Dept Psychiatry, Lab. Of Neurocognitive Develop., Pittsburgh, PA

Abstract: Dopamine is strongly associated with reward processing in the striatum, but its precise contribution to reward learning in humans has been difficult to characterize. Here, we combined behavioral (reinforcement learning) modeling with simultaneously acquired task fMRI and PET to assess the relationship of dopamine signaling and brain activation to reward related behavior.

A sample of 77 young adults (40 female, ages 18-30) were scanned in a Biograph MMR combined PET/MR scanner, during which subjects performed a rewarded map exploration task in which they attempted to accumulate rewards and learn reward probabilities for each map location. Performance data was characterized using a reinforcement learning (RL) model to assess learning parameters. A bolus/infusion paradigm was used to administer the D2/D3 ligand [^{11}C]Raclopride, and task-related DA was quantified as a change in binding potential (BP) using a modified version of the simplified reference tissue model (SRTM). Task fMRI data was acquired simultaneously, and activation was assessed by comparing BOLD responses among high, low, and no reward trials. Voxelwise analysis of the PET data across the striatum showed significant decreases in BP during task in bilateral portions of the ventral striatum (nucleus accumbens, NAcc) and dorsal putamen, indicating task-related DA release. Notably, the magnitude of DA release was greater among subjects who exhibited reward learning, compared to non-learners, in the NAcc, but not putamen. Furthermore, among learners, DA release in the NAcc was positively correlated with learning rate. DA responses were highly correlated with BOLD reward responses in the NAcc, and this effect was more closely related to parametric prediction error related activation than to reward expectation. Non-learners did not show any relationship between DA and BOLD. Our results provide direct *in vivo* support for dopamine signaling in NAcc contributing to the neural and behavioral indices of reward learning. These data confirm and extend models of reward-related dopamine signaling from rodent and primate studies.

Disclosures: **F.J. Calabro:** None. **D.F. Montez:** None. **V.P. Murty:** None. **B. Larsen:** None. **J.C. Price:** None. **B. Luna:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.24/SS4

Topic: G.02. Motivation

Support: NIH Grant F31DA042619

NIH Grant R01DA019473S1

NIH Grant R01DA019473

NIH Grant R01DA038412

Title: Exploring the mechanism by which optogenetic stimulation of ventral tegmental area dopamine neurons prevents extinction of cued approach behavior

Authors: *C. M. REYES[1], S. M. NICOLA[2]
[1]Albert Einstein Col. of Med., Bronx, NY; [2]Dept. Psychiatry, Albert Einstein Coll Med., Bronx, NY

Abstract: The nucleus accumbens (NAc) and its dopaminergic (DA) innervation from the ventral tegmental area (VTA) are involved in promoting reward-seeking behavior as well as strengthening cue-reward associations. Many NAc neurons exhibit cue-evoked excitations that are required for approach behavior elicited by a reward predictive cue. Additionally, a large body of literature suggests that DA neurons encode reward prediction errors (RPE), which serve to update the current state and alter the strength of cue-reward associations depending on the valence of the RPE (positive or negative). While RPEs presumably lead to changes in response probability, the downstream neural mechanisms from the VTA to NAc mediating this behavior remain unknown.

We hypothesized that DA neuronal activity at the predicted time of reward delivery is sufficient to reinforce cued approach behavior by maintaining the magnitude of cue-evoked excitation of NAc neurons on subsequent trials thereby blocking the transference of a negative RPE. To test this hypothesis we recorded from neurons in the NAc of *Th::Cre* rats expressing channelrhodopsin in VTA DA neurons. Animals were trained on a conditioned stimulus (CS) task in which two distinct auditory tones were presented. One tone predicted availability of a liquid sucrose reward while the other was a non-rewarded. After training, rats were subjected to an *omission* session followed by an *omission + stimulation* session. During *omission* sessions there was a 30 min baseline in the CS task followed by omission of the reward. *Omission + stimulation* sessions introduced a 20 Hz, 1s photostimulation at the predicted time of reward. During *omission* sessions we found a decrease in cue responding and a reduction in the magnitude of cue-evoked excitations of NAc neurons. Additionally, we found that stimulation of VTA DA during *omission + stimulation* sessions was sufficient to prevent extinction. Recording from NAc neurons during *omission + stimulation* sessions revealed short latency firing of NAc neurons during stimulation. In addition, the reduction in cue-evoked excitations during omission was attenuated by stimulation at the time of predicted reward. These results suggest a mechanism by which VTA DA neuronal firing influences subsequent cue-evoked excitations and thus the probability of behavioral response to the cue. Stimulation of VTA DA neurons prevents the extinction of approach behavior and our results suggest that this effect is due to a reduction in the decline in cue-evoked excitations that drive the approach response. Further experiments are underway to investigate if short latency firing is required for the maintenance of approach behavior.

Disclosures: C.M. Reyes: None. **S.M. Nicola:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.25/SS5

Topic: G.02. Motivation

Support: NIH Grant DA022340

NIH Grant DA039690

Title: Activation of ventral tegmental CB1 receptors is essential for avoidance learning

Authors: *J. M. WENZEL[1], H. M. DANTRASSY[2], E. B. OLESON[4], J. F. CHEER[3]
[1]Anat. & Neurobio., [2]Anat. and Neurobio., [3]Univ. of Maryland Sch. of Med., Baltimore, MD; [4]Neurobio. and Anat., Univ. of Colorado Denver, Denver, CO

Abstract: The mesolimbic dopamine system plays an integral role in reinforcement learning and a growing body of evidence identifies signal transduction at cannabinoid receptors as a critical modulator of this system. Utilizing an operant footshock avoidance procedure, our laboratory has previously shown that phasic activation of mesolimbic dopamine facilitates lever pressing to avoid footshock. Indeed, dopamine transients within the nucleus accumbens at the presentation of a warning signal preceding footshock delivery predict successful avoidance. Further, optogenetic activation of midbrain dopamine neurons during the warning signal enhances avoidance responding, suggesting a causal role for dopamine in avoidance. Here we examine the role of CB1 activation during learning of the operant shock avoidance task, at three time periods; early, mid and late in training. All rats were initially shaped to press a lever to terminate footshock. Once animals learned to reliably escape footshock, an avoidance contingency was introduced to the task wherein illumination of cue light served as a warning signal, which was presented 2s before the onset of footshock. Execution of a single lever press during this initial 2s interval resulted in the avoidance of footshock, whereas a lever press made after the initiation of shock delivery resulted in escape from footshock. Animals were divided into three groups to determine the effect of ventral tegmental area (VTA) CB1 receptor antagonism either from the start of training (early in training), once animals avoided footshock on ~50% of trials (mid-training), or after rats reached ~80% avoidance (late in training). Intra-VTA delivery of the CB1 antagonist rimonabant completely prevented the development of avoidance behavior when administered before each of the first 10 single daily avoidance sessions. Similarly, once rats learned to avoid footshock approximately 50% of the time, a single rimonabant administration significantly attenuated avoidance, compared to vehicle. However, rimonabant administration had no affect on avoidance behavior when delivered late in training, once rats avoided footshock on ~80% of trials. Thus, intact tegmental CB1 signaling is required for learning operant shock avoidance. However, once animals perform well on the task, CB1 activation is no longer

necessary to maintain high levels of avoidance responding. These data suggest that endogenous cannabinoid signaling exerts a previously unrecognized function in learning, whereby its effects are modulated by experience.

Disclosures: J.M. Wenzel: None. **H.M. Dantrassy:** None. **E.B. Oleson:** None. **J.F. Cheer:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.26/SS6

Topic: G.02. Motivation

Support: NIH Grant R21 DA034184

NIH Grant R01 DA09397

NIH Grant R37 EB003320

Title: Dopamine activation tracks uncertainty during operant responding

Authors: *P. MASCIA[1], J. BROWN[1], K. NESBITT[2], R. KENNEDY[2], P. VEZINA[1]
[1]Univesity of Chicago, Chicago, IL; [2]Univ. of Michigan, Ann Arbor, MI

Abstract: Pavlovian conditioning has been used in animals and humans to associate visual stimuli with different probabilities of reward. Using this approach, stimuli associated with probabilities ranging from 0 to 1.0 have been shown to differentially produce a sustained increase in dopamine (DA) activity. Stimuli associated with the most predictable outcomes (p=0 or 1.0) produce the least sustained activation while stimuli associated with the least predictable outcome (p=0.50) produce the greatest. The resulting inverted U-shaped curve has supported a relationship between sustained DA activation and uncertainty of reward. Here we describe an operant conditioning approach that supports a similar relationship between DA activation and uncertainty of reward and does so over a larger range of reward probabilities. Rats were trained extensively in 1-hr sessions to nose-poke for saccharin on escalating fixed ratio (FR) or variable ratio (VR) schedules of reinforcement using the following succession of ratios: 1, 2, 3, 5, 7, 10, 13, 16, 19, 20. The variance (a measure of uncertainty) associated with the different escalating FR ratios remained at 0 as these programmed a fixed relationship between nose-pokes and payout. On the other hand, the variance associated with the increasing VR ratios increased exponentially as these programmed an increasingly variable relationship. Remarkably, the pattern of DA overflow assessed in the nucleus accumbens with different schedules of reinforcement showed a notable similarity to these flat certainty and exponential uncertainty curves. When tested at ratios 5, 13-16, and 19-20, rats trained under the FR ratios (certain

relationship between nose-pokes and reward) showed little to no increases in DA throughout the sessions. Conversely, rats tested under the VR ratios (uncertain relationship) showed an exponential increase in DA overflow that tracked the variance of the ratios. The greatest overflow was associated with the greatest uncertainty. Notably, changes in DA overflow were not consistently associated with different behaviors (nose-pokes, dipper entries, consumption of saccharin reinforcers) and emission of the latter did not differ between FR and VR rats. These results further support a relationship between uncertainty and DA activation. Because repeated intermittent increases in DA have been implicated in the development of sensitization, these findings provide a likely mechanism for repeated exposure to conditions of uncertainty to promote the maintenance and progression of both drug and behavioral addictions such as pathological gambling.

Disclosures: P. Mascia: None. **J. Brown:** None. **K. Nesbitt:** None. **R. Kennedy:** None. **P. Vezina:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.27/SS7

Topic: G.02. Motivation

Support: NIH / NIDA Grant DA031791

NIH / NIDA Grant DA006634

Title: Differential modulation of striatal dopamine release by nicotinic receptors in adolescent and adult rats

Authors: *M. J. FERRIS, A. NORTH-FENNELL, L. L. SEXTON
Physiol. and Pharmacol., Wake Forest Sch. of Med., Winston Salem, NC

Abstract: Variability in the rate in which animals will acquire self-administration of psychostimulants, including nicotine, is a preclinical model for vulnerability to abuse drugs in humans. The variability in animals' acquisition rate can be predicted by individual differences in animals' locomotor response to a novel environment. Therefore, we investigated whether the propensity to explore a novel environment might also predict individual differences in how nicotinic acetylcholine receptors (nAChR) located on dopamine terminals in the core of the nucleus accumbens modulate rapid dopamine signals. We chose nAChRs given their well-documented ability to modulate rapid dopamine signals that are critical for acquisition behavior. Finally, we investigated whether nAChR modulation of dopamine release is similar in adolescent vs. adult rats. We assessed locomotor activity in an inescapable novel environment followed by

measurements of dopamine release using voltammetry in brain slices from adolescent (PD 28-35) and adult (PD 75-80) rats. We found no relationship between the animals' locomotor response to a novel environment and the magnitude of electrically-stimulated dopamine release in the nucleus accumbens of either cohort. In fact, no relationship between these parameters was found when dopamine was elicited under single-pulse conditions and under multiple-pulse conditions across a range of frequencies (5 Hz - 100 Hz). Following assessment of dopamine release, mecamylamine, DHβE, α-conotoxin PIA, and nicotine was bath applied to different slices from the same animal. Only adult animals with low response to a novel environment (LR) showed inhibition in dopamine release to nAChR blockade or desensitization under single pulse and low frequency, multiple-pulse stimulations that reflect tonic firing of dopamine neurons. All adolescent animals regardless of locomotor activity and adult animals with a high response to a novel environment (HR) showed greater facilitation of dopamine release to nAChR blockade or desensitization under multiple-pulse, high-frequency conditions that reflect phasic firing of dopamine neurons. Thus, LR adult animals are more sensitive to the dopamine inhibitory effects of nAChR blockade under tonic firing-like conditions while adolescent and adult HR animals are more sensitive to the dopamine facilitative effect of nAChR blockade under phasic firing-like conditions. The increased sensitivity to nAChR induced facilitation of dopamine signals elicited by phasic-like stimulation parameters may be mechanistically linked to faster and more robust acquisition of drugs of abuse and vulnerability to nicotine addiction.

Disclosures: M.J. Ferris: None. **A. North-Fennell:** None. **L.L. Sexton:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.28/SS8

Topic: G.02. Motivation

Support: COBRE 5-27132

NIH 6-36522

Title: Direct vs. indirect pathway optogenetic stimulation during conditional olfactory learning in mice

Authors: *K. TAM[1], I. OZDEN[2], E. LEE[3], J. PERGE[3], Z. YU[2], A. NURMIKKO[2], W. ASAAD[3]
[1]Mol. Pharmacology, Physiol. and Biotech., [2]Sch. of Engin., [3]Neurosci., Brown Univ., Providence, RI

Abstract: The basal ganglia has been implicated in motivational behavior, reinforcement learning, reward seeking, and action selection. The dual nature of these behaviors, i.e.

encouragement vs. discouragement (motivation), reward vs. punishment (reinforcement learning, reward seeking), go vs. no-go (action selection), has been hypothesized to be represented by two anatomically distinct pathways within the basal ganglia circuit, namely the D1 direct (striatonigral) and the D2 indirect (striatopallidal) pathways, respectively. While there have been a variety of behavioral studies supporting this hypothesis, it is still not known what specific signals are carried by each pathway. To address this issue, we studied the effects of optogenetic excitation of direct or indirect pathway during two behavioral paradigms in mice, i.e. during open-field exploration and an odor-based Go/No-Go task. For excitation of direct and indirect pathways, we injected the virus AAV5-EF1a-DIO- hChR2(H134R)-EYFP into dorsomedial striatum bilaterally in D1-Cre and A2a-Cre mice, respectively. About 3 weeks after injections two optical fibers were implanted at the injected regions.
In the open-field exploration task, mice were left in a rectangular chamber to move freely. Optogenetic stimulation was applied only when the mouse entered a predetermined region of the chamber. Our results showed that D1 mice preferred to spend more time on the stimulated region whereas D2 mice on the non-stimulated. These results are consistent with previously published work and confirm the basic functionality of our optogenetic model.
Using our verified model, we then examined the effects of optogenetic stimulation of direct and indirect pathways on learning by way of the Go/No-Go task, i.e. the speed of learning, the success rate after learning, and the reaction times. We trained mice to identify four odors delivered through an odor port. Two of the odors were used to instruct reward delivery through a water port (Go trial) and the remaining two to wait and not go to the water port (No-Go trial). One of each Go and No-Go odors were primed with optogenetic stimulation during feedback and reward period. Our data showed that neither the speed of learning, nor final task performance (>75% success rate) was significantly altered by optogenetic stimulation in D1 or D2 mice. However, we observed that the latency in initiating the subsequent trial was increased with stimulation of the indirect pathway. Overall, the odor-based Go/No-Go task results do not support the D1/D2 pathway's involvement in reward/punishment or go/no-go signals. However, the open field results are still in accord with a motivation-based signaling.

Disclosures: K. Tam: None. **I. Ozden:** None. **E. Lee:** None. **J. Perge:** None. **Z. Yu:** None. **A. Nurmikko:** None. **W. Asaad:** None.

Poster

418. Reward: Dopamine and Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 418.29/SS9

Topic: G.02. Motivation

Title: The effects of d-amphetamine on progressive ratio responding for intracranial self-stimulation and food reinforcment

Authors: M. BAGNALL, G. BOATMAN, *W. D. KLIPEC
Dept. of Psychology and Neurosci., Drake Univ., Des Moines, IA

Abstract: Rats will respond incessantly for continuous intracranial self-stimulation (ICSS) but show rapid extinction following responding for ICSS compared to food reinforcement. Since both food reinforcement and intracranial self-stimulation (ICSS) are associated with dopamine release in the mesolimbic pathways, the different patterns of behavior raise questions about the affective or reward value of ICSS. One of the problems in comparing ICSS with food reinforcement results from deprivation as an establishing operation for food but not ICSS. Here, we have compared responding on progressive ratio schedules of reinforcement, where the response requirement progressively increased within training sessions until the animal reached the maximum fixed ratio (FRMax) before it quits responding (breakpoint). Using ICSS targeted in the medial forebrain bundle and a within subjects design, we have demonstrated the breakpoint for food reinforcement is significantly higher (about FR55-60) than for ICSS (about FR5-6) in either the food deprived state or the sated state. Furthermore, we found that administration of d-amphetamine (1, 3, and 5 mg/kg i.p.), compared to saline and no injection control tests, dramatically increased the FRMax for ICSS but decreased it for food reinforcement. Based on these findings, we are hypothesizing that high, maintained levels of synaptic dopamine produced by both dense ratio schedules of ICSS and amphetamine, leading to response perseveration, are responsible for the paradoxical effects of food and ICSS as reinforcers. More specifically, these data suggest that dopamine release, by leading to the repetition of the response that it follows, may be the physiological mechanism for Skinner's reinforcement principle, rather than a pleasure based reward mechanism.

Disclosures: M. Bagnall: None. **G. Boatman:** None. **W.D. Klipec:** None.

Poster

419. Eating Disorders

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 419.01/SS10

Topic: G.07. Other Psychiatric Disorders

Support: PAPIIT-UNAM IG 200417

CONACYT 239403

Title: Time of sucrose access affects differentially binge eating behavior and Per1 in the nucleus Accumbens

Authors: *R. I. OSNAYA[1], M. PALMA GOMEZ[2], C. ESCOBAR[2]
[2]Anat., [1]UNAM, Mexico City, Mexico

Abstract: Binge eating (BE) is characterized by the consumption of large amounts of food within a discrete interval. In rodent models highly palatable foods (fats, sugars), as well as restricted food access promote BE and result in sustained dopamine release and activation of the glutamatergic system within the nucleus Accumbens. We aimed to explore the reciprocal interaction between the circadian system and BE, therefore we exploredthe influence of time of day on the development of BE and how BE can influence the rhythm of Per1 in the nucleus Accumbens. The present study explored the effect of sucrose intake during the rest phase versus the active phase on the development of BE, the influence of restricted food access on the development of BE and its effects on circadian rhythms of general activity, clock protein expression and body core temperature. The temporal expression of dopamine D1 receptor (DR1), GluR1 subunit of AMPA receptor and the clock protein PER1 were assessed in the Nucleus Accumbens and TH in the VTA. One hundred and twelve adult Wistar rats were randomly assigned to Control conditions, Restricted access to sucrose during the day (ZT5; SUC-D) or night (ZT17; SUC-N), restricted food acces to the day (RF-D) or night (RF-N) or the combination of restricted food and sucrose access in the day or night. All animals had ad libitum access to water. The experimental protocol had a duration of 5 weeks with 6 days access to sucrose per week. RESULTS: BE was 250% stronger in the SUC-N vs SUC-D and BE to sugar was not dependent of RF. RF induced overconsumption of regular chow food, an increase in locomotor activity and temperature in anticipation and response to chow access. Food restriction combined with access to sucrose increase the anticipatory response observed in general activity and core temperature. The expression of the clock protein PER1 was increased in the groups who had acces to sucrose and RF during the day and during the night, the expression of TH was also increased in the animals with access to sucrose independently of the time in which they had access to sucrose, DR1 in nucleus Accumbens was increased in the groups who had access to sucrose at night but not in the day. GluR1 was not modified between groups in the night vs. light phase. Overall our results show that BE is influenced by the time of the day, with a higher response in the night; RF can induce anticipatory locomotor activity when it is combined with sucrose access, this may be under control of day-night variations in the expression of PER1, TH and DR1 in brain reward areas. Present data will provide a better insight of factors eliciting BE and other compulsive disorders.

Disclosures: **R.I. Osnaya:** None. **M. Palma Gomez:** None. **C. Escobar:** None.

Poster

419. Eating Disorders

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 419.02/SS11

Topic: G.07. Other Psychiatric Disorders

Support: R21MH109920

R01HL127673

Title: Behavioral and metabolic characterization of eating disorder associated HDAC4-A778-T mutant mice

Authors: *K. C. DAVIS[1], M. Z. KHAN[2], M. L. LUTTER[2], H. CUI[3]
[1]Psychiatry, Univ. of Iowa Dept. of Psychiatry, Iowa City, IA; [2]Psychiatry, Univ. of Iowa, Iowa City, IA; [3]Psychiatry, Univ. of Iowa Roy J and Lucille A Carver Col. of Med., Iowa City, IA

Abstract: While eating disorders (EDs) are thought to result from a combination of environmental and psychological stressors superimposed on genetic vulnerability, the neurobiological basis of EDs remains incompletely understood. We recently reported that a rare missense mutation in the gene encoding transcriptional repressor histone deacetylase 4 (HDAC4) is associated with the risk of developing an ED in humans. To understand the biological consequences of this missense mutation, we created transgenic mice carrying this mutation by introducing the alanine to threonine mutation at position 778 of mouse Hdac4 (corresponding to position 786 of the human protein) and performed metabolic and behavioral characterization. Male mice heterozygous for HDAC4A778T did not show any metabolic or behavioral differences. In contrast, female mice heterozygous for HDAC4A778T display several ED-related feeding and behavioral deficits depending on housing condition. Individually housed HDAC4A778T female mice exhibit reduced effortful responding for high-fat diet and compulsive grooming, whereas group-housed female mice display increased weight gain on a high-fat diet, reduced behavioral despair, and increased anxiety-like behaviors. We are now in the process of characterizing homozygous HDAC4A778T mutation carriers and performing comprehensive gene expression profiling in the brain by RNA-Seq to identify the target genes affected by the HDAC4A778T mutation. The mouse line is a novel model of ED-related behaviors and will be a valuable tool to identify potential molecular pathways underlying the neurobiological basis of EDs.

Disclosures: K.C. Davis: None. **M.Z. Khan:** None. **M.L. Lutter:** None. **H. Cui:** None.

Poster

419. Eating Disorders

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 419.03/SS12

Topic: G.07. Other Psychiatric Disorders

Support: DA033373

Title: Evaluating sucrose and saccharin value in diet-induced obese rats

Authors: *S. R. BATTEN, J. BECKMANN
Psychology, Univ. of Kentucky, Lexington, KY

Abstract: The incidence of obesity has more than doubled since 1980 in the United States. Despite this increase, the neurobehavioral mechanisms that mediate obesity are largely unknown. Although it is well known that hormonal and metabolic factors influence obesity, recent evidence suggests that obesity may also be influenced by changes in reward sensitivity and associated neurocircuitry, akin to that seen in other 'reward pathologies', like addiction. However, much of this work has relied on the use of caloric reinforcers, potentially confounding metabolic effects from those of reward. The current study seeks to isolate reward changes that may occur during the onset of diet-induced obesity. Specifically, Sprague Dawley rats were trained to baseline on an operant procedure that measures the essential value of reinforcers through economic demand analyses. Baseline results show that rats value sucrose more than saccharin. Following baseline training, rats were fed a low-fat or high-fat diet for 8 weeks. Following the 8-week diet, animals fed the high-fat diet were split into 2 groups based upon weight distribution: obesity prone (top 1/3) and obesity resistant (bottom 1/3). After the split, sucrose and saccharin essential value were reassessed. The results suggest that high-fat diet produced a specific increase in sucrose and saccharin essential value for obesity-prone rats, relative to both obesity-resistant and low-fat controls. The results illustrate that a behavioral economic approach is useful in isolating the effects of diet-induced changes in reward value, and it suggests that changes in reward processes are a prominent factor in the onset of diet-induced obesity.

Disclosures: S.R. Batten: None. **J. Beckmann:** None.

Poster

419. Eating Disorders

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 419.04/SS13

Topic: G.07. Other Psychiatric Disorders

Support: NIH Grant R21MH109920

NIH Grant R01HL127673

Title: Identification of transcriptional activity driven by estrogen-related receptor alpha as a novel pathway associated with eating disorder

Authors: *H. CUI[1], K. SAITO[2], K. C. DAVIS[2], E. RODRIGUEZ CRUZ[2], B. TOTH[2], M. LUTTER[1]
[1]Psychiatry, [2]Pharmacol., Univ. of Iowa Roy J and Lucille A Carver Col. of Med., Iowa City, IA

Abstract: Eating disorders (EDs) feature serious disturbances in eating behavior and body weight regulation that are associated with a wide range of adverse psychological, physical, and social consequences. Yet, the etiology and pathogenesis of these disorders are not fully understood. We have recently identified a rare missense mutation in Estrogen-related receptor alpha (Esrra) that increases the risk of developing an ED. Interestingly, conventional Esrra-null mice display reduced body weight and decreased energy intake when fed palatable high-fat diet (HFD). However, little is known about the detailed functions of Esrra in the brain. To better understand the physiological function of Esrra in the brain, multifaceted approaches were undertaken. Immunohistochemistry confirmed a wide-spread expression of Esrra in mouse brain, which was significantly upregulated by caloric restriction. Electrophysiological recording revealed that Esrra-null female mice exhibit altered miniature excitatory postsynaptic currents on GABAergic medium spiny neurons in the ventral striatum, a well-known brain region for reward processing. Behaviorally, the Esrra-null mice display a reduced motivation for palatable HFD. Surprisingly, homozygous and heterozygous deletion of Esrra from glutamatergic and GABAergic neurons, respectively, reveal an opposite role of Esrra in excitatory and inhibitory neurons in terms of body weight homeostasis and food intake. The findings shed light on a previously unappreciated role of Esrra in the brain that may confer risk for development of an ED.

Disclosures: H. Cui: None. **K. Saito:** None. **K.C. Davis:** None. **E. Rodriguez Cruz:** None. **B. Toth:** None. **M. Lutter:** None.

Poster

419. Eating Disorders

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 419.05/SS14

Topic: G.07. Other Psychiatric Disorders

Support: R01 DA006886-19

1F31DK104645-01

Title: Accumbens processing of a food associated cue in a rat model of binge eating

Authors: *J. STAMOS, A. PAWLAK, M. SICHERER, R. PAN, C. STAMOS, J. KULIK, N. BEACHER, N. BELLO, M. WEST
Rutgers Robert Wood Johnson Med. Sch., Piscataway, NJ

Abstract: Project Summary:
Binge Eating Disorder (BED) and Bulimia Nervosa (BN) are complex disorders that involve social behavioral and body image components. BN is characterized by repeated episodes of loss

of control over eating followed by unreasonable compensatory behavior. BED is characterized by similar episodes of loss of control over food intake. However there are no unreasonable compensatory behaviors. In addition BED is associated with feelings of diminished self-worth and negative affect. In recent years there has been growing interest in the Nucleus Accumbens (Nac) involvement in eating disorders such as BED and BN. Due to their complex nature a comprehensive animal model of these disorders may not be possible. In this study we use an established rat model of binge eating (BE), which is a key component of both BN and BED, in order to study the contribution of BE behavior to disease pathology. In particular we examine how a history of BE affects Nac processing of a food associated cue. Female Sprague Dawley rats were divided into Binge Eating (BE) and Chow Control (CC) groups (n=9 per group). BE animals were subjected to a six week binge eating pretreatment. This treatment consisted of twice weekly 30 min sessions were BE animals were allowed unlimited access to a mixture of Crisco and sugar (10% sucrose by weight). Following treatment all animals had a 16 microwire array implanted into their Nac. Following recovery from surgery all animals were maintained on the binge regimen and subjected to 10 consecutive daily pavlovian trials where a tone CS was pared with a sucrose US. Each session comprised 56 trials with a variable interval of on average one minute. Behavioral measures such as number of missed trials and latency to head entry were measured over days. Neural data were divided into three phases for analysis: cue processing, approach, and consumption. Neural data from each phase were compared with analogous non-cued behavior. Furthermore, ultrasonic vocalizations (USV) were recorded during each pavlovian session and were compared to USVs recorded in a baseline session. USVs can be used to give an accurate estimation of a rat's affective state and in this context were used to see if a history of binge eating would affect the animal' affective state. Preliminary results indicate that there was an increase in the magnitude of the firing rate in the BE animals in response to the tone CS. Also the change in the magnitude of response to the CS was greater in Nac shell than Nac core in the BE but not CC animals.

Disclosures: **J. Stamos:** None. **A. Pawlak:** None. **M. Sicherer:** None. **R. Pan:** None. **C. Stamos:** None. **J. Kulik:** None. **N. Beacher:** None. **N. Bello:** None. **M. West:** None.

Poster

419. Eating Disorders

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 419.06/SS15

Topic: G.07. Other Psychiatric Disorders

Support: PAPIIT-UNAM IG-200417

CONACyT 239403

Title: Sleep deprivation induces signs of compulsive eating behavior to high fat diet associated with changes in reward brain structures

Authors: *E. N. ESPITIA, C. ESCOBAR
UNAM, Ciudad DE Mexico, Mexico

Abstract: Compulsive Eating Behavior (CEB) is characterized by eating a large quantity of palatable food (rich in fat and/ or sugar) associated with signs of anticipation, progressive escalating consumption, food-directed effort behaviors and binge eating. Sugar and fat are digested and absorptive differently, therefore we proposed that the development of CEB for both components is different. CEB is also related with activation of reward brain structures, and chronic exposition to palatable food causes ΔFOSB accumulation. Sleep deprivation increases signs of hunger and increases food intake. We aimed to explore the differential changes caused by restricted access to a high sugar diet (HSD) or high fat diet (HFD) on the development of CEB. We also explored the influence of sleep deprivation in the susceptibility to develop CEB with both type of diets. The neuronal activation measured by c-Fos and ΔFOSB accumulation was compared in reward brain structures. Rats were housed in a 12:12 h light-dark cycle with chow and water *ad-libitum*. Locomotor activity, body weight and food intake were recorded every week. Rats had restricted access to palatable diets daily for 1 hour during 4 weeks and rats were manteined one more week without diet (abstinence). In the 4th week and abstinence, all indicators of CEB were evaluated. Brains were collected to determine c-Fos activation and the accumulation of ΔFOSB in the nucleus accumbens (Acc) and prefrontal cortex (PFCx). In the HFD group binge eating and anticipation to the 1 h access started in the first week of exposure, while the HSD group started both behaviors in the 3rd week. Both groups developed effort behaviors to obtain the diet, as well as progressive consumption of the diet along the 4 weeks. The c-Fos activation in nucleus accumbens was increased in the HFD group and HFD and HSD ΔFOSB was increased in the Acc. Sleep deprivation promoted binge eating, progressive escalating consumption and high-motivated behaviors for fat, but not for sugar. Anticipation did not occur in these groups. Sleep deprivation does not impaired c-fos activation in Acc neither in PFCx, while ΔFOSB accumulation was higher in the Acc and PFCx from those who were not sleep-deprived. We conclude that HFD induces stronger signs of CEB than HSD and sleep deprivation potentiates signs of CEB only in the HFD group.

Disclosures: **E.N. Espitia:** None. **C. Escobar:** None.

Poster

419. Eating Disorders

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 419.07/SS16

Topic: G.07. Other Psychiatric Disorders

Support: NIMH Grant MH096777

NIMH Grant MH103436

Title: Taste reward prediction error signaling in adolescent anorexia nervosa

Authors: *M. DEGUZMAN, M. SHOTT, G. K. FRANK
Psychiatry, Univ. of Colorado Anschutz Med. Campus, Aurora, CO

Abstract: Introduction: Anorexia nervosa is a psychiatric disorder associated with severe low weight and increased mortality rate, yet its underlying neurobiology is not well understood. Recently, we showed that adolescents with anorexia nervosa have elevated brain response in a monetary reward paradigm associated with brain dopamine function. Here, we tested whether adolescents with anorexia nervosa (aAN) also have heightened response to a taste stimulus. Methods: We recruited forty underweight aAN (mean age =16.7±2.5 years, mean BMI =15.9±.9 kg/m^2) and thirty-five healthy weight control adolescents (mean age=15.9±2.5 years, mean BMI =21.1±1.8 kg/m^2). While undergoing functional magnetic resonance brain imaging (fMRI), all subjects learned to associate unique visual stimuli with sweet taste stimuli. Violating learned associations evoked a prediction error (PE), which has been linked with neuronal dopamine response. PE was computationally modeled based on trial sequence and regressed with brain activation. Images were preprocessed in SPM12, mean parameter estimates were extracted from regions of interest using MarsBaR, and statistical group differences were analyzed in SPSS24. Results: In response to unexpected reward receipt, aAN showed greater activation compared to control adolescents in anterior cingulum, substantia nigra, caudate, and insula. aAN showed greater PE signal than control adolescents in caudate and insula regions. Conclusions: These results align with previous findings in adults and suggest the brain reward system in aAN has heightened PE signaling, potentially due to altered dopaminergic function. Similar results using taste or monetary stimuli suggest a generalized alteration of this reward system. Further studies are needed to test whether this brain circuitry can be used to predict treatment outcome or develop therapeutic interventions.

Disclosures: M. Deguzman: None. **M. Shott:** None. **G.K. Frank:** None.

Poster

419. Eating Disorders

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 419.08/SS17

Topic: G.07. Other Psychiatric Disorders

Support: NIH Grant DA 003906

NIH Grant DA 12513

Fyssen Foundation

Title: Accumbens mechanisms for cued sucrose seeking

Authors: *A.-C. BOBADILLA, C. GARCIA-KELLER, J. A. HEINSBROEK, M. SCOFIELD, V. CHAREUNSOUK, C. MONFORTON, P. W. KALIVAS
Dept. of Neurosci., Med. Univ. of South Carolina, Charleston, SC

Abstract: The last 20 years of research supports a perspective that addictive drugs usurp brain circuits used by natural rewards, especially for the dopamine-dependent reinforcing qualities of both drugs and natural rewards. Interestingly, comorbidity between substance use disorders and binge-eating disorders is frequent; the prevalence of lifetime substance use disorders among individuals with binge eating disorders reaches 40.4% in males, and both disorders share common characteristics, like compulsive behavior, binging and distress (Schreiber, Odlaug et al., 2013). Reinstated drug seeking in animal models of relapse relies on potentiated glutamate release from cortical terminals synapsing in the nucleus accumbens core (NAcore) to stimulate metabotropic glutamate receptor5 (mGluR5) on neuronal nitric oxide synthase (nNOS) interneurons (Smith et al. 2014). In contrast to shared dopamine release, reinstated sucrose seeking does not induce glutamate spillover (Gipson et al., 2003). We hypothesized that pharmacologically promoting glutamate spillover in the NAcore would mimic cocaine-induced adaptations and potentiate cued reinstatement of sucrose seeking. We first targeted the astroglial glutamate transporter GLT-1, one of the main regulators of glutamate clearance at the synapse. Blocking GLT-1 transporter to induce glutamate spillover had no effect on sucrose seeking. However, blocking glutamate release-regulating metabotropic glutamate receptor2/3 (mGluR2/3) with selective antagonist LY 341495 potentiated cue-induced sucrose seeking. Although potentiated sucrose reinstatement following mGluR2/3 blockade was reversed by antagonizing mGluR5, direct stimulation of mGluR5 did not affect seeking. Moreover, blocking nNOS activity didn’t prevent the LY 341495-induced potentiation of sucrose seeking. Nonetheless, selective chemogenetic activation of nNOS interneurons in the NAcore potentiated cue-induced sucrose seeking. These data indicate that dysregulated presynaptic mGluR2/3 signaling is a possible site of shared signaling in drug seeking and potentiated sucrose seeking, but that, in contrast to addictive drugs, glutamate spillover due to down-regulated glutamate transport and subsequent activation of nitric oxide synthesis cannot be recruited to promote sucrose seeking.

Disclosures: A. Bobadilla: None. **C. Garcia-Keller:** None. **J.A. Heinsbroek:** None. **M. Scofield:** None. **V. Chareunsouk:** None. **C. Monforton:** None. **P.W. Kalivas:** None.

Poster

419. Eating Disorders

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 419.09/SS18

Topic: F.10. Food Intake and Energy Balance

Title: Ghrelin inhibition of the M-current in KNDy neurons and the impact of 17b-estradiol

Authors: *K. M. CONDE[1,2], T. A. ROEPKE[3]
[2]Dept. of Neurosci. and Cell Biol., [1]Rutgers Univ., New Brunswick, NJ; [3]Animal Sci., Rutgers, The State Univ. of New Jersey, New Brunswick, NJ

Abstract: Dysregulation of the hypothalamic-pituitary-gonadal (HPG) axis is caused by obesity and anorexia and both conditions negatively impact on reproduction (irregular menses, infertility, and miscarriages). The gut peptide, ghrelin, potentially mediates the influences of negative energy states (hunger, fasting) on the neuroendocrine control of reproduction. When empty, the stomach produces ghrelin to drive hunger and initiate feeding behaviors via its cognate receptor, growth hormone secretagogue receptor (GHSR). GHSR is expressed in many neurons known to regulate feeding behavior and reproduction including hypothalamic arcuate (ARC) Kisspeptin/Neurokinin B (*Tac2*)/Dynorphin (KNDy) neurons. Ghrelin signaling in other ARC neurons inhibits the M- current produced by KCNQ (Kv7) channel subunits. We hypothesize that ghrelin's control of KNDy neurons involves targeting the M-current for a dual control of reproduction and energy homeostasis. To test our hypothesis, we utilized intact Tac2-GFP adult female mice to perform whole-cell patch-clamp recordings to elicit the M-current in KNDy neurons using standard activation and deactivation protocols in voltage clamp. We first characterized the activity of the M-current in KNDy neurons using the selective KCNQ channel blocker XE991 (40 μM), which decreased the maximum peak current by 26.4 ± 5.3 pA (*n*=5). Ghrelin perfusion (100 nM) for 10 min suppressed the maximum peak current by 22.4 ± 5.1 pA (*n*=7). Additionally, we have previously reported that 17β-estradiol (E2) increases *Ghsr* expression in KNDy neurons by 6-fold and increases the M-current in NPY neurons. Subsequent experiments will characterize the impact of E2 on the KNDy M-current and ghrelin sensitivity in ovariectomized adult female mice to establish the interaction of E2 and ghrelin signaling in modulating the activity of KNDy neurons.

Disclosures: K.M. Conde: None. **T.A. Roepke:** None.

Poster

419. Eating Disorders

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 419.10/SS19

Topic: F.10. Food Intake and Energy Balance

Support: VYTP Large Grant Award

UVa Pharmacological Sciences Training Grant

Title: Medial prefrontal cortex VIP neuron activation suppresses binge-like feeding and interest in novel stimuli in the absence of an effect on food intake

Authors: *B. A. NEWMYER, M. M. SCOTT
Univ. of Virginia, Charlottesville, VA

Abstract: Novelty can be powerful driver of hyperphagia and binge eating: animals sated on one diet cease consumption if presented the same diet but will continue consumption if offered a novel palatable food source, a phenomenon known as sensory-specific satiety. Various central mechanisms driving this reward-seeking behavior synergize within the prefrontal cortex (PFC), and although considerable research exists demonstrating roles for the PFC in hedonistic food intake and food reward valuation, much less has focused on elucidating the upstream mechanisms of PFC modulation. Interneurons expressing vasoactive intestinal peptide (VIP) function as the predominant disinhibitory regulators of PFC pyramidal cells, suggesting a prominent role for this cell population in governing PFC-regulated behaviors. Thus, we sought to elucidate roles for this neuronal population in food reward seeking behavior and non-contingent food intake. We found that increasing mPFC VIP output using a Cre recombinase-dependant stabilized step-function opsin (SSFO) significantly attenuated binge consumption of highly palatable food in transgenic mice expressing Cre recombinase from the VIP peptide locus during the first exposure to this diet, while this stimulation had no effect on subsequent exposures. We also found that SSFO activation of mPFC VIP neurons significantly decreased social interaction behavior as well as novel object investigation. These findings together suggest a role for mPFC VIP neurons in novelty-based reward valuation and provide mechanistic insight into the role of this population in food intake regulation.

Disclosures: B.A. Newmyer: None. **M.M. Scott:** None.

Poster

419. Eating Disorders

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 419.11/SS20

Topic: F.10. Food Intake and Energy Balance

Support: R01DK092587 (HM)

P20GM103629 (HM)

P30DK072476 (HM)

T32-DK064584 (EQC)

Title: Contributions of lateral hypothalamic galanin signaling to feeding and affective behavior

Authors: *E. QUALLS-CREEKMORE[1], M. FRANCOIS[1], A. BRUCE-KELLER[1], C. D. MORRISON[1], H. MUNZBERG[2]
[1]Pennington Biomed. Res. Ctr., Baton Rouge, LA; [2]Pennington Biomed Res. Ctr., Baton Rouge, LA

Abstract: The lateral hypothalamus (LHA) is an important regulator of both feeding behavior and affective states. Activation of GABAergic neurons in the LHA induces robust consummatory and appetitive behaviors. Other research found hypocretin (Hcrt) neurons, also located in the LHA, regulates stress response via activation of the HPA axis and that this response is blunted by leptin signaling from non-Hcrt neurons in the LHA. We recently identified LHA galanin neurons that are non-Hcrt neurons, but innervate Hcrt neurons and modulate motivated feeding via leptin, and further suggests that LHA galanin neurons may also significantly contribute to the regulation of affective behaviors. Indeed we show that a large population of LHA galanin neurons are GABAergic and synthetic activation of LHA galanin neurons (using AAV-hM3Dq and CNO) increases appetitive feeding while also robustly decreasing stress-induced digging behaviors in the marble burying test. Importantly, activation of LHA GABA neurons maintained increases in consummatory and appetitive feeding, but stress-induced digging was conversely increased, suggesting that increased affective behavior was mediated by GABAergic neurons distinct from LHA galanin neurons. To understand if the anxiolytic effect of LHA galanin neurons also involves galanin signaling we synthetically activated LHA galanin neurons chemogenetically in the absence of intact galanin receptor 1 (GalR1) signaling pathway (GalR1KO mice) and tested for operant feeding behavior and anxiety-related behavior in the open field test, elevated plus maze, and marble burying test. We showed earlier that activation of LHA galanin neurons significantly increased operant responding and decreased anxiety-like behavior in all tested paradigms. Importantly, anxiolytic effects were not accompanied by reduced arousal indicated by a modest increase in locomotor activity in the open field test. Preliminary data confirmed that activation of LHA galanin neurons in GalR1-WT mice again promoted anxiolytic behavior in the elevated plus maze and the absence of GalR1 was not able to suppress this anxiolytic effect. Even though analysis is still ongoing, these preliminary data suggest that GalR1 may not be required for the anxiolytic effect. This data suggests LHA galanin neurons and potentially galanin signaling may represent a potential target for the treatment of disorders which present with both feeding and affective disruptions such as anorexia nervosa.

Disclosures: E. Qualls-Creekmore: None. **M. Francois:** None. **A. Bruce-Keller:** None. **C.D. Morrison:** None. **H. Munzberg:** None.

Poster

419. Eating Disorders

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 419.12/SS21

Topic: F.10. Food Intake and Energy Balance

Support: Duke Institute for Brain Sciences

Deparrtmental support

Title: Adolescents show conditioned food aversion: A strategy to study disordered eating?

Authors: E. BURNETTE, G. OCAMPO, R. WANDER, Q. WALKER, N. ZUCKER, *C. M. KUHN
Dept of Pharmacol. and Cancer Biol., Duke Univ. Hosp., Durham, NC

Abstract: Anorexia Nervosa (AN) is the leading cause of mortality due to psychiatric causes. Current animal models focus on anorexia associated with food restriction, extreme stress and or excess physical activity and no current animal model captures the key characteristics of visceral hypersensitivity leading to learned food avoidance, adolescent onset and female dominance. The present study developed a food aversion task that involved no food deprivation to investigate the developmental and sex specificity of conditioned food avoidance (CFA) in adolescent (PN 28-30) and adult (PN 60-62) rats (N = 8-16/group). Rats were placed in novel feeding cage and allowed to eat a novel cereal (Cheerios, Ch) for one hour. Twenty-four hours later, they were injected with saline or LiCl (19 mg/kg) and were allowed to eat a novel, palatable food (Froot Loops, FL). Pica was quantitated in a subset of animals for 1 hr. as a measure of nausea. Twenty four hours later, rats were given the opportunity to eat FL and another novel cereal, Apple Jacks (AJ). Cereal intake (g/kg bw) were quantitated for each cereal. Results were analyzed by 3-way ANOVA (drug x age x sex). LiCl caused pica that was greater in adults than adolescents ($p < .002$ effect of drug, $p < .002$ age x dose). Adolescents ate more of a novel food (Ch) than adults ($p < .001$ for effect of age, $p < .0002$ age x sex). All animals showed marked CFA to FL ($p < .001$ effect of drug), and adolescents showed more CFA than adults ($p < .003$ effect of age, $p < .007$ age x sex x treatment). Furthermore, females generalized the CFA to include a novel food not previously paired with LiCl more than males (AJ) $p < .001$ effect of drug, ($p < .0002$ effect of age, $p < .002$ effect of sex, and there was a trend for greater generalization in adolescents ($p < ;06$ for age x treatment). The results contrast markedly with reports from this laboratory and others of lower adolescent sensitivity in standard conditioned taste aversion tasks, and suggest that this approach may prove useful in characterizing the neural mechanisms responsible for the development of disordered eating in adolescents.

Disclosures: E. Burnette: None. **G. Ocampo:** None. **R. Wander:** None. **Q. Walker:** None. **N. Zucker:** None. **C.M. Kuhn:** None.

Poster

419. Eating Disorders

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 419.13/SS22

Topic: F.10. Food Intake and Energy Balance

Support: NIH R21 DA038504 to LCD

NIH T32 DA031115 to CPF

Title: Sex-specific alterations in dopamine transporter function from food restriction and/or exercise are amplified during adolescence

Authors: *T. L. GILMAN, W. A. OWENS, L. METZEL, C. M. GEORGE, L. C. DAWS
Cell. & Integrative Physiol., UT Hlth. Sci. Ctr. at San Antonio, San Antonio, TX

Abstract: Afflicting at least 3 percent of teenagers, eating disorders such as anorexia nervosa, bulimia nervosa, and binge eating disorder entail severe health consequences in addition to their psychological toll. Yet no effective treatments for eating disorders exist. An established regulator of both eating behaviors and physical activity, the dopaminergic system undergoes a sensitive maturation period during adolescence. Eating disorders, which are 2.5-fold more prevalent in females vs. males, typically emerge during adolescence. However, studies into the role of the dopaminergic system in ontogeny of eating disorders are lacking, as are investigations into the adolescent maturation of the dopaminergic system in females. Here we measured function of the dopamine transporter (DAT), a critical regulator of dopaminergic signaling, using both in vivo high-speed chronoamperometry and locomotor assays of cocaine response. Using an activity-based anorexia paradigm we explored how food restriction, exercise on a running wheel, or the combination thereof impacted DAT function in adult (postnatal day 90) and adolescent (postnatal day 30) Sprague-Dawley rats of both sexes. Broadly, rats exposed to both food restriction and free exercise showed leftward shifts in the dose-response to the locomotor response to cocaine, regardless of sex or age. We further observed enhanced locomotion in response to cocaine and heightened striatal function of DAT in females compared to males, and in adolescents compared to adults. Similarly, free running wheel access and restricted food access produced dramatic reductions in adolescent, but not adult, DAT-mediated dopamine uptake. Adolescent females in particular exhibit the most pronounced exercise- and diet-induced decreases in striatal DAT function. Together, these findings suggest that adolescent plasticity of the dopaminergic system confers – particularly in females – vulnerability to eating disorders and persistence of associated unhealthy behaviors (e.g., compulsive exercise). Therefore, drugs that enhance dopamine uptake or otherwise reduce dopamine signaling duration may prove efficacious in the treatment of adolescent eating disorders. Ongoing experiments are evaluating striatal DAT expression, as well as circulating insulin and leptin levels, as both of these metabolic hormones are known to

influence DAT function. Future experiments will examine how adolescent changes in DAT function and sensitivity, as a result of exercise and/or food restriction, affect voluntary access to drugs of abuse or highly palatable foods.

Disclosures: T.L. Gilman: None. **W.A. Owens:** None. **L. Metzel:** None. **C.M. George:** None. **L.C. Daws:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.01/SS23

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH Grant RO1DA034116

NIH Grant UH2NS096833

Title: Mechanisms specific to methamphetamine-associated memory disruption by nonmuscle myosin II inhibition

Authors: *M. HAFENBREIDEL[1], S. B. BRIGGS[1], S. KHAN[2], M. D. CAMERON[2], G. RUMBAUGH[3], C. A. MILLER[1]
[1]Neuroscience; Mol. Med., [2]Mol. Med., [3]Neurosci., The Scripps Res. Inst., Jupiter, FL

Abstract: Drug-associated memories, which can trigger relapse, are persistent, long-lasting, and thought to be maintained by learning-induced dendritic spine changes. During learning, dendritic spines undergo actin-dependent structural plasticity, which is then stabilized to maintain concurrent functional changes. We previously reported that depolymerizing actin with the nonmuscle myosin II inhibitor Blebbistatin (Blebb) systemically or in the amygdala (AMY) disrupts methamphetamine (METH)-associated memories. However, similar treatment does not affect cocaine (COC)-associated memories in a condition place preference (CPP) paradigm. One possibility for this difference is the longer half-life of METH, compared to COC, which suggests that longer exposure to COC (i.e. via self-administration) might render the memory susceptible. Therefore, we first assessed the ability of Blebb to disrupt context-induced reinstatement of COC seeking. Consistent with our previous results, intra-AMY Blebb had no effect on context-induced reinstatement of COC seeking. However, a home cage injection of COC (10mg/kg) paired with Blebb, with the goal of briefly reactivating actin dynamics concurrent with NMII inhibition, appeared to diminish context-induced reinstatement when tested on a subsequent day. To further follow up on the possible contribution of a longer half-life to the selective susceptibility of METH memories, we next assessed brain clearance rates of METH and COC with the goal of mimicking METH's clearance rate with COC. Previous work has shown that METH is cleared

slower following behavioral sensitization than following an acute injection. This suggests that previous drug exposure may affect future clearance rates. Therefore, we first determined the clearance rate of METH (2mg/kg) and COC (15mg/kg) on the final day of CPP training. Using mass spectrometry, we found that temporal lobe METH concentrations were approximately 60% higher than COC 15 min post-injection, despite delivering a much higher COC dose. This is consistent with the known function of the N-methyl group on METH to enhance blood brain barrier penetration. As expected, METH cleared slowly, reaching zero by ~8 hrs post-injection. Alternatively, COC concentrations dropped by more than 50% from 15 to 30 min post-injection, and were near zero by ~2 hrs. Programmable mini pumps are now being used to precisely mimic the decay rate of METH with COC in a CPP paradigm. Determining the mechanism that makes METH-associated memories uniquely vulnerable to Blebb disruption may allow for expansion to other drug-associated memories, thus allowing for further application of Blebb's therapeutic potential.

Disclosures: M. Hafenbreidel: None. **S.B. Briggs:** None. **S. Khan:** None. **M.D. Cameron:** None. **G. Rumbaugh:** None. **C.A. Miller:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.02/SS24

Topic: G.08. Drugs of Abuse and Addiction

Support: R01DA034116

R01DA034116-03S1

Title: Determining the selectivity of actin depolymerization for disrupting METH-associated memories

Authors: *S. B. BRIGGS, E. YOUNG, M. HAFENBREIDEL, G. RUMBAUGH, C. A. MILLER
Neurosci., The Scripps Res. Institute-Fl Campus, Jupiter, FL

Abstract: We have previously demonstrated that depolymerizing actin in the amygdala (AMY), either directly with latrunculin A (Lat A) or indirectly with the nonmuscle myosin II inhibitor (NMIIi) blebbistatin (blebb), produces a long-lasting and retrieval-independent disruption of the storage of memories associated with methamphetamine (METH) and amphetamine after a single treatment. Thus, depolymerizing actin, particularly through NMIIi, may prove to be a powerful therapeutic approach for targeting relapse-inducing associations. However, because the memories are being targeted in storage, it is important that the effect be selective, so as to not

disrupt other, non-drug memories. To this end, we have established that actin depolymerization delivered prior to testing has no influence on the storage of memories for fear, food reward, cocaine or morphine. In more recent work, we have sought to test the limits of the selectivity of actin depolymerization on the storage of memories that are formed either close in time or at the exact time of METH exposure in the same animal. To achieve this, we first determined the effect of depolymerizing actin on two memories that were formed over consecutive days. We found that when an auditory fear memory was learned prior to a METH-associated memory, Lat A and Blebb treatment had no effect on the fear memory. To more stringently test the selectivity of actin depolymerization for METH associations, an acute METH injection was combined with fear conditioning (FC) to pair two unconditioned stimuli with the auditory cue - METH reward and foot shock. Consistent with selective disruption of METH associations seen in other experiments, the deficits in freezing seen in animals given METH during FC and vehicle at testing was restored to the elevated levels seen in saline-treated FC that received LatA or Blebb. This result is consistent with the notion that the METH association was selectively excised, leaving the fear association intact. Taken together, these data add to the growing evidence of NMIIi's high degree of specificity for METH-associated memories.

Disclosures: S.B. Briggs: None. **E. Young:** None. **M. Hafenbreidel:** None. **G. Rumbaugh:** None. **C.A. Miller:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.03/SS25

Topic: G.08. Drugs of Abuse and Addiction

Title: BDNF mediates pervasive habit learning following withdrawal

Authors: *E. HARVEY[1], N. ANGELILLIS[2], C. AHN[3], P. J. KENNEDY[1]
[1]Psychology, [2]MIMG, [3]Neurosci., Univ. of California Los Angeles, Los Angeles, CA

Abstract: Research in the field of learning and memory suggests that the brain is composed of multiple memory systems that process and store information concurrently and may act competitively for control over learned behaviors. Among these systems are the hippocampus (HC) and dosrsomedial striatum (DMS), which have been shown to encode information flexibly to support goal-directed behaviors. Another system, the dorsolateral striatum (DLS), encodes information inflexibly and supports behaviors that are "hard-wired" (habits). Previous research from our lab and others suggest that drugs of abuse bias memory function such that the DLS predominates new behavioral learning and selection, even outside of drug-seeking contexts. The neural mechanisms supporting memory system bias following withdrawal from drugs of abuse remains largely unknown. Here we report that following extended withdrawal from chronic

cocaine, rats show a bias towards the use of DLS-dependent learning strategies whereas drug naïve controls demonstrate learning dependent on the HC. We further show that changes in behavioral learning are associated with bidirectional modulation of genes implicated in neuronal plasticity in the DLS and HC. Specifically, we identify brain derived neurotrophic factor (BDNF) as a critical regulator of memory system bias following withdrawal from cocaine and provide evidence for transcriptionally permissive and repressive chromatin states in the DLS and HC, respectively. We further show that viral-mediated overexpression of BDNF in the HC or peripheral treatment with the kappa opioid receptor antagonist (JDTic) throughout cocaine withdrawal restores HC-dependent learning. Our data suggest that chronic cocaine exposure causes lasting changes in transcriptional regulation in DLS and HC memory circuits to support the pervasive use of inflexible, "habit" learning *in lieu* of flexible, goal-directed learning. Ongoing experiments will use chromatin immunoprecitation (ChIP) to directly examine chromatin mechanisms of gene regulation in the DLS and HC following cocaine withdrawal and further investigate mechanisms of JDTic intervention strategies.

Disclosures: **E. Harvey:** None. **N. Angelillis:** None. **C. Ahn:** None. **P.J. Kennedy:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.04/SS26

Topic: G.08. Drugs of Abuse and Addiction

Support: T32 Training Grant - Translational Neuroscience of Drug Addiction

Title: Differential regulation of EGR transcription factors in hippocampus following extended access to cocaine self-administration and withdrawal

Authors: *A. GOLD[1], P. J. KENNEDY[2]
[1]Psychology, UCLA, Los Angeles, CA; [2]Psychology, Univ. of California Los Angeles, Los Angeles, CA

Abstract: Accumulating evidence suggests that the transition from recreational drug use to addiction is supported by altered plasticity in distributed brain circuits mediating reward, motivation, decision-making and learning and memory. Early growth response (EGR) family proteins have previously been implicated in a number of learning and memory processes and are thought to be critically involved in the formation of new long-term memories. Using a rat model of extended access cocaine self-administration, we report an increase in gene expression of the EGR family transcription-regulatory factors in the dorsal hippocampus (HC) compared to drug naïve and restricted access controls. These changes are further associated with decreased expression of several histone deacetylases (HDACs), suggesting that cocaine-induced changes in

HC chromatin structure may support activated gene regulation and the strengthening of memories associated with drug-taking. We further show these effects to be temporally specific as expression levels of both EGRs and HDACs return to baseline levels following a 10-day withdrawal period. Our data suggest a cocaine-mediated transcriptional regulatory mechanism that may facilitate hippocampal function and plasticity to strengthen drug-associated memories and contribute to the relapsing nature of drug addiction. Ongoing experiments are investigating the critical role of EGR family proteins in mediating the influence of context/spatial cues in the formation and maintenance of drug-associated memories and relapse.

Disclosures: A. Gold: None. **P.J. Kennedy:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.05/SS27

Topic: G.08. Drugs of Abuse and Addiction

Support: NIDA IRP/NIH

Title: Fos-expressing neuronal ensembles within the infralimbic cortex mediate extinction of cocaine seeking

Authors: *B. L. WARREN, V. SELVAM, M. VENNIRO, M. MENDOZA, F. R. SOTO DEL VALLE, D. CAPRIOLI, Y. SHAHAM, B. T. HOPE
Natl. Inst. on Drug Abuse Intramural Res. Program, Baltimore, MD

Abstract: Background: Extinction training weakens operant responding established during previous self-administration training. Extinction memories are thought to be stored within specific patterns of sparsely distributed patterns of neurons called neuronal ensembles. We previously showed that Fos-expressing neuronal ensembles in the infralimbic cortex play a role in extinction of food seeking. We now hypothesize that similar neuronal ensembles may play a role in extinction of cocaine seeking.
Methods: We trained food-restricted Long-Evans rats to lever press for infusions of cocaine for 14 days (3 h/day). On days 1-7 of self-administration, rats self-administered 1.0 mg/kg/infusion. On days 8-14, we halved the dose so that rats self-administered 0.5 mg/kg/infusion. We subsequently exposed rats to 0, 2, or 7 daily extinction sessions to assess Fos expression at three different time points following extinction training. The next day, we exposed all rats to a short 30 min test under non-reinforced conditions and assessed Fos immunoreactivity in the infralimbic cortex. We then used Daun02 inactivation in Fos-LacZ transgenic rats to test whether these Fos-expressing neurons are necessary for extinction recall.
Results: Rats increased their lever pressing for cocaine infusions over the course of self-

administration training. Furthermore, rats doubled their lever pressing when the dose of cocaine per infusion was cut in half. On test day, rats decreased their lever pressing following both 2 and 7 prior extinction sessions. We found maximal Fos expression in the infralimbic cortex of rats exposed to 2 prior extinction sessions. Daun02 inactivation of Fos-expressing neuronal ensembles associated with either self-administration or extinction recall decreased or increased lever presses compared to controls, respectively.
Conclusions: Taken together, these data suggest that neuronal ensembles within the vmPFC mediate recall of extinction of cocaine seeking in rats.

Disclosures: B.L. Warren: None. **V. Selvam:** None. **M. Venniro:** None. **M. Mendoza:** None. **F.R. Soto Del Valle:** None. **D. Caprioli:** None. **Y. Shaham:** None. **B.T. Hope:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.06/SS28

Topic: G.08. Drugs of Abuse and Addiction

Support: NIDA/NIH

Title: Whole brain Fos mapping during relapse to food seeking

Authors: *L. E. KOMER[1], R. MADANGOPAL[2], C. HEINS[3], S. A. GOLDEN[4], V. KASHTELYAN[5], C. MEJIAS-APONTE[6], Y. SHAHAM[7], B. T. HOPE[8]
[1]NIDA, Baltimore, MD; [2]Natl. Inst. On Drug Abuse IRP, Baltimore, MD; [3]Natl. Inst. on Drug Abuse, NIH, Baltimore, MD; [4]Natl. Inst. on Drug Abuse, Baltimore, MD; [5]Biomed. Res. Ctr., Natl. Inst. On Drug Abuse, Baltimore, MD; [6]Neuronal Networks Section, INRB, Natl. Inst. on Drug Abuse Intramural Res. Program, Baltimore, MD; [7]IRP/NIDA/NIH, Baltimore, MD; [8]Behav Neurosci, NIH/NIDA, Baltimore, MD

Abstract: Expression of immediate early genes (IEGs) are routinely used as an indicator of strongly activated neurons in rodent behavioral procedures. Our lab and others have shown that neuronal ensembles, identified using the activity marker Fos, mediate relapse to food and drug seeking. To investigate brain-wide circuitry that was activated during behavior, we adapted a whole brain clearing and immunolabeling technique, called iDISCO+, to generate whole brain Fos maps in mice during relapse to food seeking.
We trained mice to lever press for a palatable food pellet reward for seven days. We then tested the mice for relapse to non-reinforced food seeking (30 min test sessions) after 1 or 15 abstinence days in homecage. We transferred these mice (test group) to their homecages immediately after the test session and perfused them 60 min later; we also perfused no-test mice (homecage group) at the same time. We modified the recently published iDISCO+ method for

uniform labeling of the proteins Fos and the general neuronal marker NeuN. We optimized acquisition parameters to image immunolabeled brains using light sheet fluorescent microscopy. We observed reliable food self-administration from the mice, as well as robust relapse to food seeking during both early and late abstinence. We are currently acquiring whole brain Fos data from the experimental brains and optimizing parameters to use the open-source analysis package ClearMap for data analysis. We will quantify Fos expression by brain area, generate distribution maps, and analyze Fos activation patterns across the whole brain.
*Financial support provided by NIDA/NIH.

Disclosures: **L.E. Komer:** None. **R. Madangopal:** None. **C. Heins:** None. **S.A. Golden:** None. **V. Kashtelyan:** None. **C. Mejias-Aponte:** None. **Y. Shaham:** None. **B.T. Hope:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.07/SS29

Topic: G.08. Drugs of Abuse and Addiction

Support: NIDA IRP / NIH

Title: Examination of the role of prelimbic cortex neuronal ensembles in encoding reinstatement of palatable food seeking in rats using *In vivo* calcium imaging

Authors: *R. MADANGOPAL, C. HEINS, D. CAPRIOLI, B. LIANG, G. BARBERA, L. KOMER, J. BOSSERT, B. T. HOPE, Y. SHAHAM, D.-T. LIN
Natl. Inst. On Drug Abuse IRP, Baltimore, MD

Abstract: *In vivo* calcium imaging of awake behaving rats provides a dynamic, spatio-temporal view of task-related ensemble activity during the course of learning. Previous studies using the rat reinstatement model have implicated prelimbic cortex in reinstatement of palatable food seeking induced by non-contingent pellet priming. However, the underlying neuronal ensemble activity in prelimbic cortex that encodes pellet priming-induced reinstatement is unknown. To this end, we developed a miniature epifluorescent microscope and optimized surgical and behavioral procedures for long-term calcium imaging in prelimbic cortex of awake behaving rats.
We trained food-restricted rats to lever press for a palatable food pellet reward (FR1 reinforcement schedule; 3 pellet reward delivered after presentation of a 10-s discrete cue) in a modified trial-based design (100 trials/session; 20-s lever availability/trial; variable inter-trial interval). Next, we extinguished the rats' lever pressing in the presence of the discrete cue. We then tested the rats under extinction conditions for pellet priming-induced reinstatement (1 priming pellet delivered/trial preceding lever presentation) of food seeking. We observed reliable

food self-administration and extinction of food-reinforced responding in the trial-based procedure. We also observed robust pellet-primed reinstatement under extinction conditions. We are currently recording task-specific *in vivo* neuronal activity in prelimbic cortex during our trial-based task to identify and monitor neuronal ensembles that encode for different phases of food-seeking behavior.

Disclosures: R. Madangopal: None. **C. Heins:** None. **D. Caprioli:** None. **B. Liang:** None. **G. Barbera:** None. **L. Komer:** None. **J. Bossert:** None. **B.T. Hope:** None. **Y. Shaham:** None. **D. Lin:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.08/SS30

Topic: G.08. Drugs of Abuse and Addiction

Support: NIDA Intramural Research Program, NIH

Title: Distinct Fos-expressing neuronal ensembles in the prelimbic cortex mediate cue-induced reinstatement of cocaine and heroin seeking

Authors: *F. RUBIO, R. QUINTANA-FELICIANO, B. L. WARREN, X. LI, K. F. R. WITONSKY, Y. SHAHAM, B. T. HOPE
Behavioral Neurosci. Res. Branch, NIDA IRP, NIH, Baltimore, MD

Abstract: Learned associations between discrete cues (or contexts) and drug effects are thought to be encoded by sparsely distributed patterns of neurons called “neuronal ensembles”. Here, we determined whether cocaine and heroin seeking are encoded by distinct neuronal ensembles within the prelimbic cortex, an area in which increased the number of Fos-expressing neurons was associated with both cue-induced reinstatement of cocaine and heroin seeking.
We train *Fos-lacZ* transgenic rats to self-administer cocaine (1 mg/kg/infusion) and heroin (0.03 mg/kg/infusion) on alternating days (3-h/day; 18-days). During training, the rats were trained to lever press for cocaine or heroin with drug infusions paired with either a 3.5-s continuous or intermittent light cue (counterbalanced). After training, rats receive 10 days of operant extinction training (3-h/day) with both active levers present, but without cues or drugs.
On induction day, the rats were assigned to four groups based on the order of drug-related cues presented during induction day and test day: Cocaine-Cocaine, Heroin-Heroin, Cocaine-Heroin and Heroin-Cocaine. On induction day, the rats lever pressed for 30 min for one of the drug-related cues. Sixty minutes later, we injected the prodrug Daun02 into the prelimbic cortex which reacts with beta-galactosidase protein in the most strongly activated neurons and ablates these neurons. Two days after the induction day, we assess the effects of inactivating the cocaine- or

heroin-reward neuronal ensembles on cocaine or heroin seeking. We will present the data from this study at the meeting.

Disclosures: F. Rubio: None. **R. Quintana-Feliciano:** None. **B.L. Warren:** None. **X. Li:** None. **K.F.R. Witonsky:** None. **Y. Shaham:** None. **B.T. Hope:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.09/SS31

Topic: G.08. Drugs of Abuse and Addiction

Support: NIDA IRP

Title: sing neuronal ensembles and non-ensembles in rat prelimbic cortex following operant learning

Authors: *L. R. WHITAKER[1], B. L. WARREN[1], M. VENNIRO[2], T. C. HARTE[4], K. B. MCPHERSON[4], J. M. BOSSERT[5], Y. SHAHAM[6], A. BONCI[3], B. T. HOPE[7]
[1]Natl. Inst. on Drug Abuse Intramural Res. Program, Baltimore, MD; [3]Office of the Scientific Director, [2]Natl. Inst. On Drug Abuse, Baltimore, MD; [4]Natl. Inst. on Drug Abuse, Baltimore, MD; [5]Behavioral Neurosci., NIH, NIDA, IRP, Baltimore, MD; [6]IRP/NIDA/NIH, Baltimore, MD; [7]Behav Neurosci, NIH/NIDA, Baltimore, MD

Abstract: Learned associations between environmental stimuli and rewards drive goal-directed learning and motivated behavior. The prelimbic cortex (PLC) is known to play a role in this type of learning, although there is debate as to its specific role. Learned associations are thought to be encoded by specific patterns of sparsely distributed neurons called neuronal ensembles that are selectively activated by reward-predictive stimuli. We identify these ensembles using the neural activity marker, Fos. We hypothesized that PLC Fos-expressing neuronal ensembles play a causal role in the expression of operant learning. To test this hypothesis, we first trained rats to self-administer palatable food pellets for 1-hr/day over the course of ten days. We observed Fos expression in PLC following this operant training procedure. We then used the Daun02 inactivation method in Fos-LacZ rats and found that selective ablation of PLC neurons that were active during food self-administration training decreased lever pressing during a 30-min test session, suggesting that PLC neuronal ensembles play a causal role in the expression of operant learning. Additionally, the fundamental question of how ensemble neurons are functionally altered during learning, and which of these changes encode learned associations is unknown. To address this question, we examined intrinsic excitability in PLC Fos-expressing ensembles that were selectively activated during food self-administration training in rats. We used Fos-GFP transgenic rats to identify activated ensemble neurons (FosGFP+) and non-activated (or weakly

activated) non-ensemble neurons (FosGFP-) in an *ex vivo* brain slice preparation. Using whole cell recordings of layer V pyramidal neurons, we found increased excitability of FosGFP+ neurons and decreased excitability of FosGFP- neurons following operant training. The increased excitability of FosGFP+ neurons was driven by an increase in steady-state input resistance. The decreased excitability of FosGFP- neurons was driven by an increased contribution of small conductance calcium-activated potassium (SK) channels. To determine if the increased contribution of the SK channel was critical to retrieval of food self-administration memory, we injected the specific SK channel antagonist apamin into PLC and found no effect on lever pressing. Overall, operant training induced bidirectional plasticity in PLC neuronal ensembles and non-ensembles via distinct mechanisms. The increased excitability of Fos-expressing ensembles in PLC may play a role in encoding the self-administration memory, but not the decreased excitability in the surrounding non-ensemble neurons.

Disclosures: L.R. Whitaker: None. **B.L. Warren:** None. **M. Venniro:** None. **T.C. Harte:** None. **K.B. McPherson:** None. **J.M. Bossert:** None. **Y. Shaham:** None. **A. Bonci:** None. **B.T. Hope:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.10/SS32

Topic: G.08. Drugs of Abuse and Addiction

Support: NIDA Intramural Research Program, NIH

Title: Cue-induced reinstatement of cocaine or heroin seeking activates Fos-expressing neuronal ensembles composed of glutamatergic and GABAergic cell types in rat prelimbic cortex

Authors: *R. QUINTANA FELICIANO, F. RUBIO, X. LI, K. F. R. WITONSKY, Y. SHAHAM, B. T. HOPE
Natl. Inst. On Drug Abuse, NIH, Baltimore, MD

Abstract: Learned associations between environmental cues and drugs of abuse play an important role in inducing drug craving and relapse. In recent years, we have demonstrated that neuronal ensembles expressing the neural activity marker *Fos* play a causal role in these behaviors. Here we characterized the cellular phenotypes of *Fos*-expressing neurons in the prelimbic cortex following cue-induced reinstatement of either cocaine or heroin seeking.
For behavior, 28 Long Evans rats (females and males) were trained to self-administer cocaine (1.0 mg/kg/infusion) or heroin (0.03 mg/kg/infusion) on alternating days (9 days per drug). In 3-h daily sessions, the rats were trained to lever-press for one of the drugs; drug infusions were paired with either a 20-s continuous or intermittent light cue. Next, the rats underwent 9 3-h daily

sessions of operant extinction training in the absence of the drug cues. We observed similar levels of lever pressing during cocaine and heroin training and extinction sessions. On the reinstatement test day, two groups of rats were presented with either the cocaine or the heroin-paired cue for 60 min with both heroin- and cocaine-paired levers present. A control group of rats underwent an additional extinction session without the drug cues. Results for the test day showed selective cue-induced reinstatement in both the cocaine cue and heroin cue groups. No increase in level presses was observed in the extinction group.
For *Fos* protein expression, one group of rats was processed for immunohistochemistry 30 min after the 60-min test session. Total numbers of *Fos*-immunoreactive nuclei in prelimbic cortex for both cocaine cue (186±33 nuclei/mm^2) and heroin cue (258±46 nuclei/mm^2) groups were significantly higher than that in the extinction group (74±18 nuclei/mm^2). For double-labeling of *Fos* and cell type markers, a separate group of rats was processed for RNAscope *in situ* hybridization. In rats that reinstated lever pressing for either cocaine or heroin cues, preliminary results indicate that >80% of *Fos*-expressing neurons co-expressed the vesicular glutamate transporter (*Vglut*) while <15% co-expressed the vesicular gamma-aminobutyric acid transporter (*Vgat*).
Overall, we found that cue-induced reinstatement of cocaine or heroin seeking activated prelimbic cortex *Fos*-expressing neuronal ensembles with both glutamate and GABA cell types.

Disclosures: R. Quintana Feliciano: None. **F. Rubio:** None. **X. Li:** None. **K.F.R. Witonsky:** None. **Y. Shaham:** None. **B.T. Hope:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.11/SS33

Topic: G.08. Drugs of Abuse and Addiction

Support: European Union, co-financed by the European Social Fund (EFOP-3.6.1.-16-2016-00004)

European Union, co-financed by the European Social FunPécs University, Medical School (PTE ÁOK KA-2015-15)

Title: The role of oxytocin and dopamine interaction in amygdaloid reinforcing mechanisms and anxiety

Authors: *K. LASZLO[1,2], K. FITTLER[3,2], T. OLLMANN[3,2], A. KOVACS[3,2], O. ZAGORACZ[3,2], L. PECZELY[3,2], E. KERTES[3,2], Z. KARADI[3,2,4], L. LENARD[3,2,4]
[1]Inst. of Physiol., Pécs University, Med. Sch., Pecs, Hungary; [2]Ctr. of Neuroscience, Univ. of

Pecs, Pecs, Hungary; [3]Inst. of Physiology, Univ. of Pécs, Med. Sch., Pecs, Hungary; [4]Mol. Endocrinol. and Neurophysiol. Res. Group, Pecs, Hungary

Abstract: Neuropeptide oxytocin (OT) is involved in the regulation of social and non-social behavior. The central nucleus of the amygdala (CeA), part of the limbic system, plays an important role in learning, memory, anxiety and reinforcing mechanisms. CeA has been shown to be rich in OT-receptors. Our previous findings indicated that in the rat CeA OT has a dose dependent positive reinforcing and anxiolytic effect. The aim of our present study was to examine in the CeA the possible effects of OT and dopamine (DA) D2 receptor antagonist sulpiride on reinforcement in place preference test and on anxiety in elevated plus maze test. Male wistar rats were microinjected bilaterally with 10 ng OT (Sigma: O6379, injected in volume of 0.4 µl). In different group of animals 4 µg DA D2 receptor antagonist (sulpiride: Sigma: S7771 dissolved in sterile saline, injected in volume of 0.4 µl) was applied. Other animals received DA D2 receptor antagonist 15 min before 10 ng OT treatment or vehicle solution into the CeA.
Rats receiving 10 ng OT spent significantly longer time in the treatment quadrant during the test session in conditioned place preference test. Prior treatment with DA D2 receptor antagonist blocked the rewarding effects of OT. Antagonist in itself did not influence the time rats spent in the treatment quadrant. In elevated plus maze test, rats receiving 10 ng OT spent significantly longer time on the open arms. Prior treatment with DA D2 receptor antagonist blocked the effects of OT.
Our results show that in the rat CeA OT has positive reinforcing and anxiolytic effects. DA system plays a role in positive reinforcing and anxiolytic effects of OT because DA D2 receptor antagonist can block these actions.

Disclosures: K. Laszlo: None. **K. Fittler:** None. **T. Ollmann:** None. **A. Kovacs:** None. **O. Zagoracz:** None. **L. Peczely:** None. **E. Kertes:** None. **Z. Karadi:** None. **L. Lenard:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.12/SS34

Topic: G.08. Drugs of Abuse and Addiction

Support: National Institute of Mental Health Intramural Research Funding: 1ZIAMH002386

Title: NCS-Rapgef2, the protein product of the neuronal Rapgef2 gene, is a specific activator of D1 dopamine receptor-dependent ERK phosphorylation in mouse brain

Authors: *S. Z. JIANG, W. XU, S. SWEAT, M. EIDEN, C. GERFEN, A. EMERY, L. EIDEN
NIMH/NIH, Bethesda, MD

Abstract: The neuritogenic cyclic AMP sensor (NCS) is encoded by the *Rapgef2* gene. This protein links cyclic AMP elevation to the activation of extracellular signal-regulated kinases 1/2 (ERK) in neurons and neuroendocrine cells (Emery et al., Sci. Signaling 6 (281), ra51, 2013; J. Biol. Chem. 289: 10126, 2014). Cyclic AMP elevation does not cause ERK activation in the human embryonic kidney 293 (HEK293) cells, which are devoid of Rapgef2 protein. Transducing these cells with a vector encoding *Rapgef2* reconstituted cAMP-dependent ERK activation. Mutagenesis of Rapgef2 revealed that its cyclic nucleotide binding domain (CNBD) is necessary for cAMP-dependent activation of ERK. A point mutation of one CNBD residue abrogated cAMP-ERK coupling, while deletion of the CNBD yielded a constitutively active protein. In vivo, RT-PCR analysis revealed two classes of transcripts from the *Rapgef2* gene: one was found in all tissues and one was specific to neuronal and endocrine tissues. In adult mice, expression of the full-length ~167KD Rapgef2 protein (NCS-Rapgef2) was evident only in neuronal and neuroendocrine tissues, corresponding to the expression pattern of the neuronal/neuroendocrine-specific transcript, initiated at an alternative first exon (termed by us exon 1'). In the brain, NCS-Rapgef2 is expressed specially in neurons, with prominent expression in corticolimbic excitatory neurons and striatal medium spiny neurons (MSNs). The expression of NCS-Rapgef2 in the nucleus accumbens (NAc) suggested the possibility of engagement of NCS-Rapgef2 via the dopamine D1 receptor. Rapgef2 dependent ERK activation was confirmed in neuroendocrine NS-1 cells expressing the human DRD1a receptor: treatment with D1 agonist SKF81297 (10 µM) caused cAMP-dependent ERK activation and neuritogenesis, while deletion of the *Rapgef2* gene abolished this effect. In vivo, ERK phosphorylation induced by SKF81297 (2 or 5 mg/kg, ip) was significantly attenuated in the hippocampal dentate gyrus and basolateral amygdala of CamK2α-$Cre^{+/-}$; $Rapgef2^{cko/cko}$ mice, which have forebrain-specific NCS-Rapgef2 ablation. Likewise, SKF81297-induced ERK phosphorylation in NAc MSNs was significantly reduced in $Rapgef2^{cko/cko}$ mice injected with AAV-Synapsin-Cre. Treatment with psychostimulants cocaine (30 mg/kg, i.p.) or amphetamine (10 mg/kg, i.p.) significantly increased ERK phosphorylation in NAc MSNs, which was also blocked by prior injection of AAV-Synapsin-cre. We conclude that D1-dependent ERK phosphorylation in mouse brain requires NCS-Rapgef2 expression, and therefore that ERK-dependent behaviors mediated by the D1 receptor are likely to be NCS-Rapgef2-dependent.

Disclosures: S.Z. Jiang: None. **W. Xu:** None. **S. Sweat:** None. **M. Eiden:** None. **C. Gerfen:** None. **A. Emery:** None. **L. Eiden:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.13/SS35

Topic: G.08. Drugs of Abuse and Addiction

Title: Regional expression of Egr2 in substructures of the reward circuit regulates distinct aspects of cocaine-induced behaviors

Authors: *D. MUKHEREE, B. GONZALES, E. ITSKOVITS, L. IZAKSON, A. ZASLAVER, A. CITRI
Hebrew Univ., Jerusalem, Israel

Abstract: Neuroadaptations underlying maladaptive behavior driven by drugs of abuse have been shown to depend on transcriptional changes in the reward circuit. We thus hypothesized that the information contained in inducible transcription could enable identification of novel molecular substrates driving long term neuroplastic changes affected by drugs of abuse. To this end, we performed high throughput time dependent transcriptional profiling of 7 brain structures in the reward circuit, in response to acute (novel), repeated or challenge (reinstatement) administrations of cocaine. Our results demonstrate that the individual cocaine experiences were characterized by distinct but consistent region specific expression of immediate early genes (IEG), with low inter-individual variability and clear temporal dynamics. Comparing between the experience-induced transcriptomes, we observed the most significant changes in spatiotemporal patterns of expression of *Egr2*, a transcription factor belonging to the Early Growth Response family. Moreover, using machine learning algorithms for supervised classification, we have identified that this region specific expression of *Egr2* was a reliable marker for segregation and unique clustering of the different cocaine experiences much more accurately compared to conventional IEGs like *Arc* and *Fos*. These indications suggested that spatially defined expression of *Egr2* could be playing an important role in encoding the different aspects of a cocaine experience. To test this we performed targeted shRNA mediated knockdown of *Egr2* expression in the distinct anatomical substructures of the striatum, and subjected them to cocaine induced sensitization and place preference behavior. Initial results from such experiments indicated that expression of *Egr2* in the NAc regulates only psychomotor activation but in the DS affected both psychomotor behavior and cocaine induced place preference. In each case, knockdown of *Egr2* expression led to attenuated expression of *Arc* and *Fos* in the manipulated brain regions but no change in other brain areas. Taken together, this study provides a system level description of the spatiotemporal transcriptional architecture induced by distinct cocaine experiences. We report the identification of novel marker *Egr2* that is highly informative regarding the extent of cocaine exposure of mice, and whose region-specific expression is responsible for encoding distinct aspects of cocaine dependent behaviors. However, the underlying cellular mechanisms by which region specific expression of *Egr2* implements the encoding of cocaine induced behaviors is a topic for further investigation.

Disclosures: D. Mukheree: None. **B. Gonzales:** None. **E. Itskovits:** None. **L. Izakson:** None. **A. Zaslaver:** None. **A. Citri:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.14/SS36

Topic: G.08. Drugs of Abuse and Addiction

Support: ANPCyT, FONCYT, PICT 2012-0924, Argentina

ANPCyT, FONCYT, PICT 2012-1769, Argentina

ANPCyT, FONCYT, PICT 2014-2499

IBRO-PROLAB, 2016. IBRO-PROLAB Program

ANPCyT, FONCYT, PICT 2015-2594, Argentina.

Title: Retrieval of cocaine and caffeine-associated memory selectively alters early gene expression in the prefrontal cortex and nucleus accumbens of mice

Authors: J. P. PRIETO[1], J. MUÑIZ[2], B. GONZALEZ[2], J. L. CADET[3], M. SCORZA[1], F. J. URBANO[4], *V. BISAGNO[2]
[1]Inst. de Investigaciones Biologicas C. Estable, Montevideo, Uruguay; [2]ININFA-CONICET, Buenos Aires, CABA, Argentina; [3]Mol. Neuropsychiat Br., NIH, Baltimore, MD; [4]IFIBYNE-CONICET, Buenos Aires, Argentina

Abstract: Caffeine is the world's most popular psychostimulant and is frequently used as an active adulterant in many illicit drugs including cocaine. Previous studies have shown that caffeine, acting as an adulterant, can potentiate the stimulant effects of cocaine and cocaine-induced drug seeking behavior. However, little is known about the effects of this drug combination on reward-related learning, a key process in the maintenance of addiction and vulnerability to relapse. The goal of the present study was thus to determine if caffeine can enhance cocaine-conditioned memory and lead to differential mRNA expression in the nucleus accumbens (NAc) and prefrontal cortex (PFC). Mice were treated with caffeine (5 mg/kg, CAF), cocaine (10mg/kg, COC), or their combination (caffeine 5 mg/kg + cocaine 10 mg/kg, CAF+COC) in a Conditioned Place Preference (CPP) test. NAc and PFC tissues were dissected immediately after the CPP test or after a single conditioning session for mRNA expression analysis. CAF+COC induced a marked change of preference to the drug conditioned side of the CPP and a significant increase in locomotion compared to COC. Gene expression analysis of early genes as well as dopamine and adenosine receptor subunits revealed up-regulation of D1 receptor subunit, cFos and FosB in the NAc, and cFos, EGR1 and NPAS4 in the PFC of the CAF+COC animals. With a single conditioning session, CAF+COC induced changes only in D1 receptor subunit (up-regulation) in the NAC and cFos (down-regulation) like the CAF group,

whereas in the PFC, we observed increased expression of D1, D2 and A1 receptor and cFos in both CAF and CAF+COC groups. Interestingly, we found that NPAS4 gene expression was increased only in the PFC of the CAF+COC. Our study provides evidence that caffeine acting as an adulterant is capable of potentiating reward-associated memories elicited by cocaine. This is associated with specific changes in early gene expression that were observed only in groups that received the combination of both psychostimulants in the context of CPP memory retrieval. These results highlight the importance of caffeine in the maintenance of cocaine addiction probably by neural plasticity mechanisms linked to strengthening learning associated with the drug-environment.

Disclosures: **J.P. Prieto:** None. **J. Muñiz:** None. **B. Gonzalez:** None. **J.L. Cadet:** None. **M. Scorza:** None. **F.J. Urbano:** None. **V. Bisagno:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.15/SS37

Topic: G.08. Drugs of Abuse and Addiction

Support: NIDA Grant T.J.G., DA017949

NIDA Grant T.J.G.,1U01DA041632

Jean Phillips Shibley Endowment

Penn State Biobehavioral Health Department

Title: Acute nicotine disrupts consolidation of contextual fear extinction and alters long-term memory-associated hippocampal kinases

Authors: *S. GADIWALLA[1], T. J. GOULD[1], M. G. KUTLU[1], J. TUMOLO[2], B. GARRETT[2]
[1]Penn State Univ., University Park, PA; [2]Dept. of Psychology, Neurosci., Temple Univ., Philadelphia, PA

Abstract: Long-term memory consolidation and its cell-signaling cascades are regulated by nicotinic acetylcholine receptors (nAChRs). Evidence showed that nicotine, an agonist of nAChRs, has enhanced hippocampus dependent fear learning by altering phosphorylation patterns of several cell-signaling kinases that are involved in the consolidation of long-term memories including ERK1/2 and JNK1. In addition, past work has indicated that acute nicotine impairs contextual fear extinction. However, the effects of nicotine administration on cell signaling pathways during acquisition and consolidation of fear extinction memories are unknown. In this study, we examined the effects of acute nicotine on the phosphorylation of

dorsal and ventral hippocampal ERK1/2 and JNK1 during contextual fear conditioning. Our results showed that acute nicotine administered immediately, and 30 minutes, but not 6 hours following extinction impaired contextual fear extinction. This suggests that acute nicotine injections within the memory consolidation window disrupts the consolidation of contextual fear extinction memories. We also found that acute nicotine administered prior to extinction sessions downregulated the phosphorylated forms of ERK1/2 in the ventral hippocampus, but not dorsal hippocampus, and JNK1 in the dorsal and ventral hippocampus during the consolidation phase on the 3rd day of extinction, results which were not present on the 1st extinction day. Finally, our results showed that acute nicotine injections immediately after each session upregulated the phosphorylated form of ERK1/2 in the ventral hippocampus, but did not affect dorsal hippocampal ERK1/2 phosphorylation or phosphorylated form of JNK1. In summary, these results illustrate that acute nicotine impairs contextual fear extinction potentially by altering molecular processes responsible for the consolidation of extinction memories.

Disclosures: S. Gadiwalla: None. **T.J. Gould:** None. **M.G. Kutlu:** None. **J. Tumolo:** None. **B. Garrett:** None.

Poster

420. Circuit and Molecular Mechanisms of Memory in Addiction

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 420.16/SS38

Topic: G.08. Drugs of Abuse and Addiction

Support: PSI2015-68600-P

Title: Activity changes surrounding cerebellar perineuronal nets in cocaine induced preference memories

Authors: *J. GUARQUE-CHABRERA[1], I. GIL-MIRAVET[2], M. MIQUEL[3]
[1]Psychobiology, Univ. Jaume I, Villarreal, Spain; [2]Univ. Jaume I, Castellon, Spain; [3]Jaume I Univ., Castellon, Spain

Abstract: Perineuronal nets (PNNs) are a critical part of synaptic plasticity machinery. A PNN is an aggregation of extracellular matrix molecules (versican, aggrecan, neurocan, brevican, hyaluronan, tenascin-R, link proteins, and semaphorin 3A) in a net-like manner that envelopes the perikaryon and proximal dendrites of special subsets of neurons. Since PNNs begin to form at the end of the brain development and because of the stability of its components, they have been proposed as a candidate mechanism for learning and memory storage. In the adult brain, PNNs create the conditions to restrict synaptic plasticity modifications. Given that addiction could be considered the result of pathological learning, PNNs are considered to contribute to the maintenance of drug-induced conditioned memories after prolonged drug abuse. Drug-induced

plasticity changes in PNNs in relation to addiction are receiving increasing support but still very few studies are focused on animal models of drug addiction. Previous studies from the lab showed that after the infralimbic deactivation PNNs were upregulated in the dorsal cerebellar cortex. Also, the infralimbic cortex and the dorsal cerebellum seem to be part of a functional and structural network which would work on restraining goal-directed behavior when drug-cue associations are acquired. Therefore, PNNs in this network might be an important mechanism for storage of drug-induced conditioned memories. The present research aimed to assess whether an infralimbic deactivation would affect glutamatergic activity surrounding PNNs in the dorsal granule cell layer. We used vGluT1, vGluT2 expression in order to estimate changes in neural activity. Our results indicated that in the sham group cocaine-induced preference conditioning was accompanied by an upregulation of PNN expression and a significant increase in the glutamatergic activity. However, the infralimbic deactivation reduced significantly glutamatergic activity around the fully condensed PNNs. These findings suggest that the infralimbic cortex has the capacity to regulate activity in the dorsal cerebellar cortex.

Disclosures: **J. Guarque-Chabrera:** None. **I. Gil-Miravet:** None. **M. Miquel:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.01/SS39

Topic: G.08. Drugs of Abuse and Addiction

Support: Gabriel Bosse is supported by a CIHR fellowship

Title: Modeling drug seeking using zebrafish

Authors: *G. BOSSE, R. T. PETERSON
Univ. of Utah, Col. of Pharm., Salt Lake City, UT

Abstract: America is in the midst of a major opioid addiction crisis owing to the alarming increase in opioid overdose-related fatalities. One of the main challenges is that no treatment is currently available to cure this addiction, as replacement therapies are the only viable options. Drug addiction is a state defined by compulsive engagement in naturally rewarding behavior or drug use, despite adverse consequences. The zebrafish (*Danio rerio*) has become an excellent tool to study mental health disorders, due to its physiological and genetic similarity to humans, ease of genetic manipulation, and feasibility of small molecule screening. Zebrafish have been shown to exhibit characteristics of addiction to drugs of abuse in non-contingent assays, including conditioned place preference. However, thus far, a single contingent assay has been reported for only one type of consumption abuse.
Using inexpensive electronic, mechanical, and optical components, we developed an automated

opioid self-administration assay for zebrafish. Thus, enabling us to measure drug seeking and gain insight into the underlying biological pathways. Zebrafish trained in the assay for five days exhibit robust self-administration, which is dependent on the function of the μ-opioid receptor. In addition, as with other animal models, we can also use a progressive ratio protocol to test our conditioned fish and they also continue to seek the drug despite an adverse consequence. Furthermore, fish trained in our assay also showed signs of stress and anxiety upon withdrawal of the drug. Finally, we validated our assay by confirming that self-administration in zebrafish is dependent on several of the same molecular pathways as other animal models.
Given the ease and throughput of this assay, it will enable the identification of important biological pathways regulating drug seeking and could lead to the development of new therapeutic molecules to treat addiction.

Disclosures: G. Bosse: None. **R.T. Peterson:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.02/SS40

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH/NIDA grant R21 DA037728 (Gewirtz, JC and Harris, AC, Co-PIs)

NIH/NIDA training grant T32 DA007097 (Swain, Y; Molitor T, PI)

Minneapolis Medical Research Foundation Translational Addiction Research Program (Harris, AC, PI)

Minneapolis Medical Research Foundation Career Development Award (Harris, AC, PI)

Title: Higher anhedonia during withdrawal from initial opiate exposure predicts lower levels of subsequent morphine self-administration in rats

Authors: *Y. SWAIN[1,2], P. MUELKEN[2], A. SKANSBERG[1,2], M. KRUEGER[1,2], D. MOTZ[1,2], Z. HAAVE[3,2], M. G. LESAGE[1,4,2], J. C. GEWIRTZ[1,3,2], A. C. HARRIS[1,4,2]
[1]Dept. of Psychology, Univ. of Minnesota Twin Cities, Minneapolis, MN; [2]Minneapolis Med. Res. Fndn., Minneapolis, MN; [3]Dept. of Neurosci., [4]Dept. of Med., Univ. of Minnesota, Twin Cities, Minneapolis, MN

Abstract: Understanding factors contributing to individual differences in opioid addiction vulnerability is essential for developing more effective preventions and treatments. Opiate withdrawal leads to negative affective (emotional) symptoms including anhedonia. The role of

withdrawal in individual differences in addiction vulnerability is unclear, with some findings indicating that greater withdrawal sensitivity is associated with lower addiction vulnerability and other findings indicating the opposite. The goal of the current study was to evaluate the ability of anhedonia during initial opiate withdrawal to predict individual differences in subsequent addiction vulnerability as measured using i.v. morphine self-administration (MSA) in rats. Rats were first tested for withdrawal sensitivity following acute injections of morphine (i.e., “acute dependence”), measured as elevations in intracranial self-stimulation (ICSS) thresholds (anhedonia-like behavior) during naloxone-precipitated and spontaneous withdrawal. Rats were then catheterized and allowed to acquire MSA (0.2 mg/kg/infusion, 2 hour/day sessions). Greater escalation (sensitization) of naloxone-precipitated withdrawal across repeated morphine injections and greater peak spontaneous withdrawal severity were both associated with lower infusion rates during acquisition of MSA. These data support some previous studies indicating an inverse relationship between withdrawal sensitivity and drug use propensity, and suggest that high anhedonia during withdrawal from initial opiate exposure could be protective against subsequent opiate addiction. These findings also establish a model that could be used to understand the behavioral and neurobiological mechanisms underlying individual differences in opiate addiction vulnerability.

Disclosures: **Y. Swain:** None. **P. Muelken:** None. **A. Skansberg:** None. **M. Krueger:** None. **D. Motz:** None. **Z. Haave:** None. **M.G. LeSage:** None. **J.C. Gewirtz:** None. **A.C. Harris:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.03/SS41

Topic: G.08. Drugs of Abuse and Addiction

Title: The role of the paraventricular nucleus of the thalamus in the augmentation of heroin seeking induced by chronic food restriction

Authors: *A. CHISHOLM, D. RIZZO, N. GONZALEZ, C. MCANULTY, E. FORTIN, A. BUMBU, U. SHALEV
Psychology, Concordia Univ., Montreal, QC, Canada

Abstract: Drug addiction is a chronic disorder that is characterized by compulsive drug seeking and involves switching between periods of compulsive drug use, abstinence, and relapse. In both human addicts and animal models of addiction chronic food restriction has been shown to increase rates of relapse. Previously, our laboratory has demonstrated a robust increase in drug seeking following a period of withdrawal in chronically food-restricted rats compared to sated rats. To date, the neural mechanisms that mediate the effect of chronic food restriction on drug

seeking have not been elucidated. However, the paraventricular nucleus of the thalamus (PVT) appears to be a promising candidate to investigate. The PVT is uniquely placed to contribute to both homeostatic control and drug seeking systems. Thus, the objective of the current study was to study the effect of PVT activation on heroin seeking under food restriction conditions.
Prior to heroin self-administration training, male Long Evans rats were injected with a viral vector carrying an excitatory Designer Receptor Exclusively Activated by Designer Drug (DREADD) into the PVT. Next, rats were trained to self-administer heroin over the course of 10 days (0.1 mg/kg/infusion; i.v.). Following training, rats were removed from the operant conditioning chambers and placed into drug withdrawal for 16 days. Over the withdrawal period, rats were exposed to a mild food restriction (90% of baseline body weight) or were given unrestricted access to food. On the 14^{th} and 16^{th} day of the withdrawal period, two drug-seeking tests were conducted in which rats were injected (i.p.) with either CNO (6 mg/kg), to activate the DREADDs, or vehicle, 20 minutes prior to test. Injectors' placement was verified using immunohistochemistry.
All rats reliability learned to self-administer heroin. As expected, food-restricted rats demonstrated an augmented heroin seeking during the heroin-seeking test in comparison to sated rats. Preliminary results suggest that PVT activation may reduce heroin seeking in food-restricted rats.
These results suggest that PVT activity appears to play a role in heroin seeking. Furthermore, PVT activity may modulate the augmentation of heroin seeking following chronic food restriction.

Disclosures: **A. Chisholm:** None. **D. Rizzo:** None. **N. Gonzalez:** None. **C. McAnulty:** None. **E. Fortin:** None. **A. Bumbu:** None. **U. Shalev:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.04/SS42

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH Grant DA037287

Title: The effect of NMDA antagonists on the reinforcing properties of oxycodone and nalbuphine in rhesus macaques

Authors: *K. L. NICHOLSON, K. L. SHELTON, M. L. BANKS
Pharmacology/Toxicology, Virginia Commonwealth Univ., Richmond, VA

Abstract: Therapeutic use of prescription analgesics has more than quadrupled in the last decade and concomitantly the incidence of prescription opioid abuse has also doubled. Interestingly,

some studies suggest that NMDA antagonism can decrease the reward-related effects of opioid agonists as has been shown in rodents. Using a behavioral economic approach, we examined the impact of co-administration of the NMDA antagonist ketamine on self-administration levels of the opioid medications oxycodone (high efficacy mu agonist) and nalbuphine (low efficacy mu agonist) to assess its potential as an abuse-deterrent adjunctive therapy. Adult male rhesus monkeys were surgically implanted with indwelling intravenous catheter and trained to self-administer 100 mcg/kg/infusion ketamine during daily 3-hr operant sessions under a fixed ratio (FR) 50 schedule of reinforcement. Initially, dose effect curves were determined for ketamine, oxycodone and nalbuphine alone under the following FR schedules: 10, 20, 50, 100 and 200, presented in a counterbalanced order for periods of 4 consecutive days. Subsequently two doses of oxycodone (3 or 10 mcg/kg/infusion) or nalbuphine (1 or 3 mcg/kg/infusion) were tested under the same FR schedules in combination with varying proportions of ketamine. Demand curves relating drug intake to FR and measures of reinforcement were generated for oxycodone and nalbuphine alone as well as in the presence of ketamine. Each drug alone, ketamine, nalbuphine and oxycodone, functioned as a reinforcer. Ketamine increased measures of reinforcement for oxycodone and nalbuphine. Unlike previous work utilizing pretreatments with NMDA antagonists, concurrent administration of ketamine failed to attenuate and actually increased total opioid intake at one or more doses of ketamine. These findings, combined with our previous findings in measures of antinociception (Cornelissen et al., 2017) suggest the combination of ketamine and opioids provides limited therapeutic potential either as an adjunctive analgesic or as an abuse deterrant. However, because ketamine itself has abuse liability, studies examining the effects of the unscheduled NMDA antagonist memantine in combination with each of the opioid drugs are ongoing.

Disclosures: **K.L. Nicholson:** None. **K.L. Shelton:** None. **M.L. Banks:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.05/SS43

Topic: G.08. Drugs of Abuse and Addiction

Support: ASU College of Liberal Arts and Sciences

Title: Effects of heroin intake on prosocial behavior in rats

Authors: S. E. TOMEK, G. M. STEGMANN, *M. OLIVE
Psychology, Arizona St Univ., Tempe, AZ

Abstract: Clinical studies show a negative correlation between heroin use and levels of prosocial behaviors. Laboratory rats are capable of prosocial or empathy-like behavior. For

example, under normal conditions, when a rat is placed in the vicinity of another rat that is confined to a plastic restrainer, it will release or “rescue” the confined rat. The present study utilized this paradigm to determine the effects of heroin intake on prosocial behavior in rats. For two weeks, rescuer rats were given the opportunity to release their cagemate, and the occurrence and latency to free the confined rat was recorded. After rescuing behavior was established, rats were randomly assigned to self-administer either heroin (0.06 mg/kg/infusion, i.v.), or sucrose pellets (45 mg) as a control, for 10-14 days. Next, rats were retested for rescuing behavior once daily for 3 days, during which they had a choice between freeing the trapped cagemate or continuing to self-administer their respective reinforcer. Results indicated that the proportion of rats in the sucrose group that continued to rescue their cagemate instead of self-administering sucrose was 76%, whereas the proportion of rats in the heroin group rescuing their cagemate was 0%, and these rats continued to self-administer heroin instead. Thus, rats with a history of heroin self-administration show deficits in prosocial behavior, consistent with specific diagnostic criteria for opiate use disorder. Since activity in the insular cortex is known to be correlated with both drug craving and important abilities needed for social interaction, future studies are planned to attempt to restore prosocial behavior following heroin intake using chemogenetic approaches.

Disclosures: **S.E. Tomek:** None. **G.M. Stegmann:** None. **M. Olive:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.06/SS44

Topic: G.08. Drugs of Abuse and Addiction

Support: Senior Research Career Scientist Award from the Department of Veterans Affairs

Department of Defense Grant DM090595

Louisiana Board of Regents Grant ITRS-015B

Louisiana Board of Regents LEQSF(2015-20)-GF-11

Title: Endomorphin analog ZH853 shows reduced locomotor activation and physical dependence relative to morphine

Authors: *A. T. AMGOTT-KWAN[1], T. J. HUNTER[1], A. K. FEEHAN[1], J. E. ZADINA[1,2,4,3]
[1]Neurosci. Program, Brain Inst., Tulane Univ., New Orleans, LA; [2]Med., [3]Pharmacol., Tulane Univ. Sch. of Med., New Orleans, LA; [4]SE Louisiana Veterans HCS, New Orleans, LA

Abstract: Morphine remains the gold standard for treatment of moderate and severe pain, but its clinical use is limited by severe side effects. The current opioid epidemic further emphasizes the

need for safer alternative analgesics with reduced abuse potential. Our lab previously characterized endomorphin analog ZH853 as a potent agonist of the mu-opioid receptor (1). ZH853 provides more potent or equipotent and longer-lasting analgesia as compared to morphine in neuropathic, inflammatory, post-operative, and visceral pain models (2). ZH853 also showed reduced side effects including reduced respiratory depression, motor impairment, tolerance, hyperalgesia, and glial activation (1). The transition from casual drug use to addiction is thought to be initially motivated by the rewarding properties of the drug resulting from mesolimbic dopamine activation. Continued drug use is later maintained by physical dependence and reward tolerance resulting from allostatic dysregulation (3). The reduced reward of ZH853 was recently supported by a lack of conditioned place preference and self-administration (1). Here we further demonstrate the reduced abuse liability of ZH853 by assessing locomotor activation and physical dependence. Opioid-induced locomotor activity in mice indicates mesolimbic dopaminergic activation, which is associated with reward (4). Acute subcutaneous administration of morphine, but not equi-antinociceptive doses of ZH853, induced locomotor activation in CF1 mice, suggesting differential dopaminergic activation. Chronic escalating doses morphine, but not ZH853, produce naloxone-precipitated jumping and weight loss, indicators of physical dependence. These data suggest the potential of ZH853 to provide long-term pain treatment with reduced abuse liability relative to morphine.

(1) Zadina, JE *et al.* (2016). *Neuropharmacology* **(105)**: 215-227.
(2) Feehan, AK *et al.*, (2017). *Manuscript in preparation.*
(3) Koob, GF, & Volkow, ND. (2016). *The Lancet* **(8)**: 760-773.
(4) Wise, RA, & Bozarth, MA (1987). *Psychological Review* **(94)**:469 – 492.

Disclosures: A.T. Amgott-Kwan: None. **T.J. Hunter:** None. **A.K. Feehan:** None. **J.E. Zadina:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.07/SS45

Topic: G.08. Drugs of Abuse and Addiction

Support: Supported by NIDA-IRP

Title: VK4-116, a highly selective and metabolically stable dopamine D3R antagonist, selectively inhibits oxycodone reward without compromising oxycodone's antinociceptive effects in rats

Authors: *Z.-B. YOU[1], G.-H. BI[2], V. KUMAR[2], E. L. GARDNER[2], Z.-X. XI[2], N. H. AMY[2]
[1]Behavioral Neurosci, [2]Mol. Targets and Medications Discovery Br., NIDA-IRP/NIH/DHHS, Baltimore, MD

Abstract: The recent epidemic-like increase in prescription opioid misuse and abuse is a serious challenge to public health worldwide. Exploring potential pharmacotherapies for prescription opioid abuse has become a major focus in recent drug abuse research. We have recently developed a highly selective and metabolically stable dopamine D3 receptor antagonist - VK4-116, which significantly inhibits oxycodone-induced conditioned place preference, hyperactivity and locomotor sensitization (Kumar et al., J Med Chem, 2016). However, it is unknown whether this compound is similarly effective in other oxycodone-induced addiction-like behaviors and whether it interacts with oxycodone's analgesic effects. In the present study, we found that: 1) VK4-116 pretreatment (5-25 mg/kg, i.p.; 15 min prior to testing) significantly inhibited oxycodone self-administration in rats that had already acquired self-administration behavior; 2) VK4-116 pretreatment significantly inhibited oxycodone-induced drug-seeking following behavioral extinction; 3) repeated pretreatment with VK4-116 (5, 15 mg/kg, once daily for 5 consecutive days) dose-dependently blocked acquisition of oxycodone self-administration in drug-naïve rats; 4) VK4-116 pretreatment inhibited naloxone-precipitated conditioned place aversion; and 5) VK4-116 pretreatment at doses of 5 or 15 mg/kg (i.p., 15 min before oxycodone) showed no effects on the antinociceptive action of oxycodone as assessed by hot-plate testing. At 25 mg/kg, VK4-116 significantly potentiated oxycodone's antinociceptive effects. Together, our findings indicate that VK4-116 may serve as a novel promising medication for the treatment and prevention of prescription opioid abuse with potential benefit of augmenting the analgesic effects of prescription opioids. (Supported by NIDA IRP)

Disclosures: Z. You: None. **G. Bi:** None. **V. Kumar:** None. **E.L. Gardner:** None. **Z. Xi:** None. **N.H. Amy:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.08/SS46

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH Grant R01 DA025674

NIH Grant R01 DA025674 06S1

NIH Grant R03 DA034886

Title: Preconception opiate exposure enhances the rewarding effects of cocaine in offspring

Authors: *F. M. VASSOLER, A. M. TOORIE, E. M. BYRNES
Dept. of Biomed. Sci., Tufts Univ. Cummings Sch. of Vet. Med., North Grafton, MA

Abstract: The United States is in the midst of an opiate epidemic, with abuse and misuse of prescription and illegal opioids increasing steadily over the past decade. A growing body of evidence describes that environmental exposures, such as opioids, can impact the physiology and behavior of subsequent generations. The current study was designed to test the hypothesis that maternal or paternal exposure to opioids prior to pregnancy alters abuse liability in subsequent generations. Female or male adolescent rats were administered morphine at increasing doses (5-25 mg/kg, s.c.) or saline for 10 days (P30-39). Animals then remained drug free for at least 3 weeks. During adulthood (P70-P90), animals were bred with drug-naïve partners. Male and female adult offspring (F1 animals) were tested for either morphine or cocaine self-administration acquisition, progressive ratio, extinction, and reinstatement (0.75-morphine and 0.5-cocaine mg/kg/infusion). In addition, mu-opioid receptor expression levels as well as β-endorphin peptide levels were measured in the nucleus accumbens and ventral tegmental area. There were both drug- and sex- dependent effects on all phases of the self-administration paradigm that indicate decreased morphine reward and attenuated relapse-like behavior and yet increased cocaine reward and enhanced relapse-like behavior in Mor-F1 animals compared with Sal-F1 animals. Additionally, both receptor and cognate peptide levels were altered in Mor-F1 animals. The results demonstrate that even limited opioid exposure during adolescence can have lasting effects across multiple generations.

Disclosures: F.M. Vassoler: None. **A.M. Toorie:** None. **E.M. Byrnes:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.09/SS47

Topic: G.08. Drugs of Abuse and Addiction

Support: R01 DA036582

Title: Craving and addiction severity in rat models of heroin and cocaine sequential polydrug abuse

Authors: *E. CRUMMY
Univ. of Washington, Seattle, WA

Abstract: Polydrug abuse is commonly observed in drug abusers with reports of 30% to 80% of heroin addicts also using cocaine (Leri et. al, 2003); heroin abusers all also 15x more likely to develop addictions to cocaine (National Survey on Drug Use and Health, 2011-2013). While high prevalence of concomitant and sequential drug abuse has been reported, polydrug abuse, especially sequential abuse, is relatively understudied and little is understood about the mechanisms underlying craving and addiction severity resulting from polydrug use. Initial

studies in our lab have found decreased use of cocaine in rats alternatively self-administering cocaine and heroin compared to rats self-administering cocaine alone, but consistent heroin use in polydrug users versus single users of heroin. When examining incubation of craving, however, incubation was observed in polydrug users for both heroin and cocaine, comparable to craving behavior demonstrated in single use of heroin or cocaine across ten sessions. To further characterize if cocaine and heroin use recruited different circuits, rats self-administered cocaine and heroin on alternating days for a total of eight sessions, then were either placed in their home cage or allowed to continue to self-administer cocaine for a thirty day period. Rats demonstrated craving after a thirty day withdrawal period from heroin, regardless of access to cocaine, suggesting separate circuitry is involved in craving for these drugs and consistent with previous reports of selective reinstatement for heroin or cocaine when administered priming injections (Leri & Stewart, 2001). Experiments are currently underway to further characterize the impacts of polydrug use history and prolonged exposure to drug on craving and addiction severity. Using self-administration threshold paradigms to assess maximal prices that rats are willing to take to maintain initial drug consumption levels.

Disclosures: E. Crummy: None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.10/SS48

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH Grant RO1 DA036582

Title: Striatal medium spiny neurons regulate reinstatement of heroin-seeking

Authors: *T. J. O'NEAL, S. M. FERGUSON
Neurosci., Univ. of Washington, Seattle, WA

Abstract: Lethal heroin overdoses have risen dramatically in recent years, and the difficulty faced by individuals attempting to abstain from use has exacerbated this. Indeed, nearly 90% of heroin users will relapse within 3 months of abstinence, and the likelihood of successful, long-term abstinence decreases with each relapse. Although the rewarding effects of heroin are known to act primarily through mu opioid receptor-mediated modulation of dopaminergic transmission within the cortico-basal ganglia network, the circuit perturbations that contribute to reinstatement of heroin-seeking remain poorly understood. Notably, glutamatergic projections from the ventral subiculum to the nucleus accumbens (NAc) have been shown to mediate reinstatement of heroin-seeking, but the NAc contains two homogeneously interspersed populations of GABAergic output neurons: D1 receptor-expressing medium spiny neurons (D1-MSNs) and D2 receptor-

expressing MSNs (D2-MSNs). D1-MSNs and D2-MSNs have differential downstream and local projection pathways, and the role of each cell type in heroin-seeking is not clear. To begin to address this, rats underwent surgery for implantation of indwelling jugular catheters and stereotaxic surgery for infusion of a retrograde CAV-Cre virus into either the ventral tegmental area (VTA) or ventral pallidum (VP) along with infusion of an AAV-DIO-hM4Di virus into the NAc to allow pathway-specific manipulation of D1-MSNs and D2-MSNs, respectively. Rats were trained to self-administer heroin (0.075 mg/kg/infusion) on an FR1 schedule using an intermittent-access paradigm (5 min ON, 25 min OFF; 6 hr total) for 10 days, during which drug delivery was signaled by a cue light. Following training, rats underwent extinction testing in the self-administration chambers in the absence of heroin or cue light availability. To assess the role of D1-MSNs and D2-MSNs in heroin-seeking, rats then underwent cue-induced reinstatement tests following injection of clozapine-N-oxide (CNO; 5 mg/kg) to selectively inactivate these circuits. Inhibition of D1-MSNs significantly attenuated heroin-seeking following CNO injection. Currently, the role of D2-MSNs in cued-reinstatement is under investigation, as well as the effect of stimulating D1-MSNs or D2-MSNs using an AAV-DIO-hM3Dq virus, both of which will offer additional insight into how these distinct pathways regulate heroin-seeking during abstinence.

Disclosures: T.J. O'Neal: None. **S.M. Ferguson:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.11/SS49

Topic: G.08. Drugs of Abuse and Addiction

Support: Norwegian Research Council grant #213751

Title: The contribution of 6-acetylmorphine to the rewarding effects of heroin

Authors: *A. S. KVELLO[1], F. BOIX[1], J. M. ANDERSEN[1], J. MORLAND[2], I. BOGEN[1]
[1]Dept. of Forensic Sci., Oslo Univ. Hosp., Oslo, Norway; [2]Norwegian Inst. Publ. Hlth., Oslo, Norway

Abstract: Background: The acute behavioral effect of heroin is mainly mediated by its first metabolite, 6-acetylmorphine (6-AM). However, 6-AM's implication in the rewarding properties of heroin remains uncertain. The current study aimed to examine the contribution of 6-AM to heroin reinforcement and reward. **Methods**: Male C57BL/6J mice were tested for conditioned place preference (CPP) induced by equimolar doses (1.25-5 µmol/kg) of heroin or 6-AM, administered prior to conditioning for three consecutive days. A CPP test was performed on the fourth day. The locomotor activity of the animals was recorded during each 20 min conditioning

session. To further investigate the individual contributions from heroin and 6-AM to heroin-induced reward, additional mice were pretreated with a 6-AM specific antibody (anti-6-AM mAb; 10-200 mg/kg) 24 hours before subjected to the same experimental procedure as described above, but using only a dose of 2.5 µmol/kg heroin or 6-AM. **Results**: Both heroin and 6-AM produced CPP in mice. However, heroin CPP scores (367 and 381 s) were twice as high as 6-AM CPP scores (159 and 168 s) induced by doses of 2.5 and 5 µmol/kg, respectively. Locomotor sensitization was seen after heroin and 6-AM doses of 2.5 and 5 µmol/kg, but not after 1.25 µmol/kg, and was more profound in the heroin group compared to the 6-AM group. We found no correlation between the expression of CPP and locomotor sensitization. Pretreatment with anti-6-AM mAb prior to heroin and 6-AM injections blocked the expression of both CPP and locomotor sensitization to both opioids, however, a high mAb dose (200 mg/kg) was required to block heroin-induced CPP. **Conclusions**: Altogether, our results suggest that heroin holds a higher rewarding potential compared to 6-AM, and that CPP and locomotor sensitization are two phenomena mediated through different mechanisms in the rodent brain.

Disclosures: **A.S. Kvello:** None. **F. Boix:** None. **J.M. Andersen:** None. **J. Morland:** None. **I. Bogen:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.12/SS50

Topic: G.08. Drugs of Abuse and Addiction

Title: An investigation of the reinforcing effects of diazepam and midazolam in rats trained to self-administer heroin

Authors: *S. L. SMITH[1], S. HOLLAND[1], R. A. GRAY[2], D. J. HEAL[1]
[1]Renasci Ltd, Nottingham, United Kingdom; [2]Sovereign House, Vision Park, GW Pharmaceuticals, Cambridge CB24 9BZ, United Kingdom

Abstract: Although benzodiazepines (BDZs) have reinforcing effects in humans (eg Griffiths et al, 1984, Psychopharm **84**:147-54; Braun et al, 2008, Eur Neuropsychopharmacol **18**:723-8) and are recreationally abused, very few published studies exist that describe the reinforcing effects of the BDZs in rats. We have addressed this deficiency by investigating whether diazepam and/or midazolam serve as positive reinforcers in rats trained to self-administer heroin.
Mildly food-restricted, male, Sprague-Dawley rats were initially trained to lever-press for food rewards before being surgically implanted with in-dwelling jugular catheters. Rats were allowed to self-administer heroin (15µg/kg/inj) on a fixed ratio (FR3) schedule of reinforcement in 2hr training sessions. After establishment of consistent heroin self-administration, the rats were subjected to saline extinction. The reinforcing effects of diazepam (1, 3, 4.5 or 10µg/kg/inj) and

midazolam (0.3, 1, 1.5, 2.25 or 3μg/kg/inj) were then evaluated on a FR3 schedule in 2hr sessions. In the first part of the experiment, if a BDZ served as a reinforcer (>6 inj/session) in an individual rat, a 4hr progressive ratio (PR)/break-point analysis was performed. Results are mean ± SEM.
Heroin maintained self-administration in rats (17.6±0.5 inj/session, n=39) at levels significantly greater ($p<0.001$) than saline (3.7±0.2 inj/session, n=39). Diazepam served as a positive reinforcer in 50% (4/8) and 43% (3/7) rats at 3 and 10μg/kg/inj, respectively, and midazolam in 29% (5/17) and 69% (11/16) rats at 1 and 1.5μg/kg/inj, respectively. When the group mean results were analysed diazepam (3μg/kg/inj) [7.0±2.1 inj/session, n=8] and midazolam (1.5μg/kg/inj) [7.3±1.3 inj/session, n=16] maintained self-administration at levels significantly greater than saline ($p<0.05$). The number of infusions of all doses of diazepam and midazolam were significantly ($p<0.001$) lower than heroin. The break-points for responding (mean lever-presses/inj) for diazepam were 18.9±2.8 (n=4) and 13.0±1.0 (n=3) at 3 and 10μg/kg/inj, respectively. For midazolam, they were 17.1±2.8 (n=3) and 13.2±0.7 (n=5) at 1 and 1.5μg/kg/inj, respectively. All benzodiazepine break-points were significantly ($p<0.05$) lower than heroin (39.4±6.9, n=10).
This study is the first to systematically investigate whether BDZs substitute for heroin in rats, and compare their reinforcing effects relative to heroin. Diazepam and midazolam maintained self-administration at some, but not all, doses in heroin-maintained rats indicating their reinforcing effects are weak in this species. This was confirmed by their low break-points for drug reinforcement.

Disclosures: **S.L. Smith:** A. Employment/Salary (full or part-time):; RenaSci Ltd. **S. Holland:** A. Employment/Salary (full or part-time):; RenaSci Ltd. **R.A. Gray:** A. Employment/Salary (full or part-time):; GW Pharmaceuticals. **D.J. Heal:** A. Employment/Salary (full or part-time):; RenaSci Ltd.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.13/SS51

Topic: G.08. Drugs of Abuse and Addiction

Support: NIDA Grant R01DA37207

Title: The selective M_5 negative allosteric modulator ML375 attenuates remifentanil self-administration without blocking morphine-induced analgesia in rats

Authors: ***B. GUNTER**[1,3], R. W. GOULD[1,3], M. P. MILLER[1], M. BUBSER[1,3], C. W. LINDSLEY[1,3,2], C. K. JONES[1,3]

[1]Pharmacol., [2]Chem., Vanderbilt Univ., Nashville, TN; [3]Vanderbilt Ctr. for Neurosci. Drug Discovery, Nashville, TN

Abstract: Opioid use disorder (OUD) has reached epidemic proportions in the United States. Yet, current treatment options remain limited to opioid maintenance and detoxification strategies, which are associated with high rates of relapse, abuse potential, and other adverse side effects. Accumulating evidence suggests that selective inhibition of the M_5 muscarinic acetylcholine receptor (mAChR) subtype may provide a novel approach for the treatment of OUD. M_5 is the only mAChR subtype expressed on midbrain DA cell bodies and selective activation of M_5 increases midbrain DA cell firing and DA efflux in the nucleus accumbens; effects that are blunted or absent in the M_5 knockout (KO) mice. Furthermore, M_5 KO mice exhibit reduced morphine place preference without affecting morphine-induced analgesia. Recently, we reported the successful discovery and optimization of a novel series of highly selective M_5 negative allosteric modulators (NAMs), represented by the M_5 NAM tool compound ML375. Previously, we have demonstrated that ML375 acutely attenuates both the reinforcing effects and relative reinforcing strength of cocaine. In the present studies, we determined whether ML375 would also attenuate opioid self-administration in rats without blocking opioid-induced analgesia.

Male Sprague-Dawley rats were trained to self-administer remifentanil under a fixed ratio schedule of reinforcement and the ability of ML375 to attenuate the reinforcing effects of remifentanil at each unit dose was then evaluated. The effects of ML375 were also determined when rats responded under a progressive ratio (PR) schedule for remifentanil alone and in combination with cocaine. Finally, the potential analgesic effects of ML375 alone and in combination with morphine were assessed using a hot plate apparatus.

ML375 reduced remifentanil self-administration at each dose under a FR 10 schedule. Furthermore under a PR schedule, ML375 reduced the reinforcing strength of remifentanil. Isobolographic and dose-addition analysis of the effects of ML375 on remifentanil/cocaine combinations suggests ML375 attenuates the supra-additive effects of a single remifentanil/cocaine combination. Finally, our preliminary data indicate that ML375 does not have analgesic properties when administered alone, nor does it have any effect on morphine-induced analgesia.

Taken together, these results suggest that selective negative allosteric modulation of the M_5 mAChR receptor may represent a promising novel pharmacotherapy for the treatment of OUD.

Disclosures: B. Gunter: None. **R.W. Gould:** None. **M.P. Miller:** None. **M. Bubser:** None. **C.W. Lindsley:** None. **C.K. Jones:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.14/SS52

Topic: G.08. Drugs of Abuse and Addiction

Support: NIDA/NIH

Title: Effect of blockade of mu, delta, and kappa opioid receptors on context-induced reinstatement of oxycodone seeking

Authors: *J. K. HOOTS[1], I. FREDRIKSSON[1], C. CIFANI[1,2], S. ADHIKARY[1], Y. SHAHAM[1], J. M. BOSSERT[1]
[1]Behavioral Neurosci., NIDA/NIH, Baltimore, MD; [2]Sch. of Pharm., U Camerino, Camerino, Italy

Abstract: Background: High relapse rates perpetuate prescription opioid addiction, which drives the current drug overdose epidemic in the US. This relapse often occurs after exposure to places previously associated with opioid use. Here, we describe a rat model of context-induced reinstatement of oxycodone seeking after repeated cycles of drug self-administration and extinction-induced abstinence. We also determined the role of mu, delta, and kappa opioid receptors (MOR, DOR, KOR) in this reinstatement. Methods: We trained rats to self-administer oxycodone (0.1 and 0.05 mg/kg/infusion; 7-10 days/dose, 6-h/d) in Context A; lever pressing was paired with a discrete cue. Next, we extinguished the lever pressing in the presence of the discrete cue in Context B for 8-11 days (6-h/d). We then tested the rats for reinstatement of oxycodone seeking in both contexts under extinction conditions (1-h/day). Next, we re-trained the rats to self-administer oxycodone in Context A, re-extinguished their lever pressing in Context B, and re-tested them for reinstatement in both contexts. Prior to testing, we injected (s.c) the rats with vehicle or antagonists of MOR (naltrexone; 0.5 or 1.0 mg/kg), DOR (naltrindole; 7.5 or 15 mg/kg), or KOR (LY2456302; 5 or 10 mg/kg). Results: We observed reliable context-induced reinstatement of oxycodone seeking after repeated cycles of drug self-administration and extinction. Systemic injections of naltrexone, but not naltrindole or LY2456302, decreased this reinstatement. Conclusions: We established a rat model of context-induced relapse to oxycodone seeking after repeated cycles of drug self-administration and abstinence and demonstrated a role of MOR but not KOR or DOR in this relapse.

Disclosures: J.K. Hoots: None. **I. Fredriksson:** None. **C. Cifani:** None. **S. Adhikary:** None. **Y. Shaham:** None. **J.M. Bossert:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.15/SS53

Topic: G.08. Drugs of Abuse and Addiction

Title: Cannabidiol blocks morphine place preference in mice

Authors: J. R. MARKOS[1], H. M. HARRIS[1], W. GUL[2], M. A. ELSOHLY[2], *K. J. SUFKA[1]
[1]Dept. of Psychology, [2]Res. Inst. of Pharmaceut. Sci., Univ. of Mississippi, Oxford, MS

Abstract: This study determined whether the cannabis constituent cannabidiol attenuates the development of morphine reward in the conditioned place preference paradigm. Mice received either saline or morphine in combination with increasing doses of cannabidiol using 3 sets of conditioning trials. After drug-place conditioning, morphine mice displayed robust place preference that was dose-dependently attenuated by cannabidiol. Further, the dose of cannabidiol that was effective against morphine reward was void of rewarding and aversive properties. The finding that cannabidiol blocks opioid reward suggests that this compound may be useful as an abuse deterrent.

Disclosures: J.R. Markos: None. **H.M. Harris:** None. **W. Gul:** None. **M.A. ElSohly:** None. **K.J. Sufka:** None.

Poster

421. Opioids and Behavior

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 421.16/SS54

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH Grant R37DA009815

PA DOH SAP#4100062216

Title: Time-of-day effects on heroin intake

Authors: *A. A. COFFEY, J. FANG, P. S. GRIGSON
Penn State Col. of Med., Hershey, PA

Abstract: Addiction is a chronic disease characterized by multiple relapses. In the United States, heroin abuse is increasing dramatically. Circadian rhythms govern more than just sleep, and there is mounting evidence of circadian effects on drug abuse. For example, most heroin overdose hospitalizations occur in the early morning hours. There also are circadian effects on metabolism of drugs and dopamine release/reuptake, potentially changing the rewarding properties of drugs of abuse across the 24 hour cycle. Interestingly, cocaine intake in rats is highest mid-light cycle and mid-dark cycle and lowest early light and early dark cycle. However, time-of-day effects on heroin self-administration have not been assessed in rats.
Previously, we have shown that rats self-administering heroin during their inactive light cycle reverse their sleep patterns relative to rats self-administering saline during the light cycle and rats

self-administering heroin or saline during the dark cycle. This reversal persisted for 4 days after the rats no longer had access to heroin. In the current study, we sought to investigate whether this circadian disruption affects drug intake, motivation to take drug, and relapse-like behavior. Over 18 trials, 38 male Sprague-Dawley rats were trained to self-administer heroin for 6 hours daily during the second half of either their inactive light cycle (n=17) or their active dark cycle (n=21). Motivation to take drug was tested using a progressive ratio schedule of reinforcement for one trial during the third week of acquisition. Rats were then tested in an extinction paradigm where no drug was available, followed by 13 additional days of abstinence, and an extinction/drug-induced reinstatement test. Rats with dark cycle access to heroin took more drug than those with light cycle access, but there was no difference in the motivation to take drug between the groups. Dark access rats also failed to extinguish seeking across the second extinction test, however, there was no difference between the groups in drug seeking during reinstatement. Understanding the circadian effects on drug intake has the potential to inform risk factors for worsening drug addiction and relapse, particularly if circadian disruption feeds into this process.

Disclosures: **A.A. Coffey:** None. **J. Fang:** None. **P.S. Grigson:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.01/SS55

Topic: G.08. Drugs of Abuse and Addiction

Support: NSF Grant I0S-1557755

R03DA038734

Title: *In vivo* electrochemical assessment of dopamine release during conditioned withdrawal in heroin dependent rats

Authors: *S. SCHELP, K. PULTORAK, E. OLESON
Univ. of Colorado, Denver, Denver, CO

Abstract: The centers for disease control and prevention reports that heroin abuse has doubled for Americans aged 18-25. Drugs of abuse, such as heroin, are commonly thought to increase the concentration of dopamine (DA) in the nucleus accumbens core (NAcc), although their effects on phasic DA release events remains to be fully characterized. In contrast, when an addicted subject is withdrawn from a substance such as heroin, there is a clear suppression of phasic DA activity in the NAcc. Heroin withdrawal presents characteristic symptoms, most predominately increased craving for the drug, which contributes to further abuse. While withdrawal symptoms

have been shown to be conditioned to previously neutral stimuli through classical conditioning, there is a lack of electrochemical evidence of conditioned withdrawal and its effects on non-drug related stimuli. Here, we explored how conditioned withdrawal in heroin addicted rats affects DA. We provided rats 23 hour access to IV heroin over 28 days. The 28 days are split into four blocks of increasing doses ranging from 0.06 mg/kg to 6 mg/kg to more accurately simulate the progression of heroin addiction. During the 28 day history, circadian changes in heroin, food, and water intake were altered and overall consumption of heroin increased. We then, investigated how conditioned withdrawal affects cue-evoked dopamine release in the NAcc using fast scan cyclic voltammetry (FSCV) during conditioned suppression of food maintained responding. Here, animals were first trained to respond for food under an FR1 schedule for 3 consecutive days where food availability was predicted by a cue-light placed above the lever. This was followed by 3 consecutive days of conditioned withdrawal training where a tone was conditioned to naloxone-precipitated withdrawal. On test day, animals responded for food under normal conditions, but the withdrawal associated tone was presented concurrently with cue-light onset during 33% of trials. By simultaneously presenting the withdrawal paired tone with the food predictive cue light we observed a substantial suppression of DA concentration at the cue light relative to non-paired trials. After conditioned withdrawal test day sessions, IV 6 mg/kg heroin was administered followed by 1.0 mg/kg naloxone methiodide and 1.0 mg/kg naloxone HCl during FSCV to determine if peripheral nervous system inputs contribute to withdrawal induced DA suppression. These data demonstrated that conditioned suppression of food maintained responding induced by conditioned heroin withdrawal is accompanied by a transient suppression in dopamine concentration.

Disclosures: S. Schelp: None. **K. Pultorak:** None. **E. Oleson:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.02/SS56

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH Grant DA036372

Title: Sex differences in gene expression in the amygdala in morphine dependence and withdrawal

Authors: *S. J. O'SULLIVAN[1], J. PARK[1,2], J. GORKY[1], C. C. THEISEN[3], B. A. REYES[4], E. J. VAN BOCKSTAELE[4], J. S. SCHWABER[1]
[1]Pathology, Anatomy, & Cell Biol., Thomas Jefferson Univ., Philadelphia, PA; [2]Chem. and Biomolecular Engin., Univ. of Delaware, Newark, DE; [3]Neurobio. & Anat., [4]Pharmacol. & Physiol., Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: Opioid withdrawal is characterized by dysphoria, nausea, anxiety and fear. The negative reinforcement theory of addiction postulates that avoidance of these negative physical and emotional symptoms drives drug-seeking. The amygdala plays a central role in this context as it is a limbic structure responsible for fear and anxiety that also processes autonomic inputs, generates autonomic outputs, and has consistently demonstrated involvement in addiction and motivated behaviors. The organization of gene networks that underlie amygdalar function in opioid dependence and withdrawal reveal genetic network behaviors and specific pathological drivers that contribute to the observed drug-seeking behavior via negative reinforcement. We use a combination of laser capture microdissection and microfluidic RT-qPCR to accurately assay 96 transcripts at both the single-cell and tissue level in morphine-dependent and withdrawn rats. Previous experiments at the single-cell level in the Central Nucleus of the Amygdala (CeA) demonstrated global transcriptional shifts along with cell type-specific gene alterations—especially in stress-related and inflammatory genes. We are building on these studies by analyzing putative sex differences in transcriptomes within the CeA as well as the Basolateral Nucleus of the Amygdala (BLA) following chronic morphine exposure and naltrexone-precipitated withdrawal in male and female rats. Addiction mechanisms at large, and transcriptional networks specifically, that differ between the sexes not only offer a novel view of opioid addiction, but may also provide insight into potential targets for intervention.

Disclosures: S.J. O'Sullivan: None. **J. Park:** None. **J. Gorky:** None. **C.C. Theisen:** None. **B.A. Reyes:** None. **E.J. Van Bockstaele:** None. **J.S. Schwaber:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.03/SS57

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH grant number P20GM103506NH-INBRE

Title: ΔFosB expression in human postmortem brain tissue from opioid overdoses

Authors: *L. JABBOUR, F. CARTEE, M. CAISSE, K. HAYNES
Franklin Pierce Univ., Rindge, NH

Abstract: Background: ΔFosB is a truncated form of FosB, a transcription factor from the Fos family of transcription factors. It is established that ΔFosB, normally absent in brain tissue, accumulates following conditions of prolonged exposure to drugs of abuse. It has also been shown to be up-regulated following abuse of natural rewards and under chronic stress. ΔFosB is believed to be a molecular switch for establishing long lasting changes in gene expression following abuse, however, these changes have only been described in animal models.

Objective: We hypothesize that ΔFosB has an equivalent role in the human brain. Our objective is to characterize its expression in the human brain tissue obtained at autopsy of opioid overdoses.
Methods: We obtained post-mortem brain tissue from 24 cases of opioid overdose and 4 control cases, age and sex-matched, where death was not caused by a drug overdose. The brain tissue was immediately fixed in 10% formalin and subsequently dissected. We used immunohistochemistry to evaluate *in-situ* protein expression in various regions of the mesolimbic system. We used microtubule-associated protein (MAP) and a neuronal specific nuclear protein (NeuN) as a positive control, and ran a slide with no primary antibody for every assay conducted.
Results: We observed positive staining for ΔFosB in the nucleus accumbens and more specifically, between the islands of Calleja. We have also tested amygdala but at time of abstract submission, we had seen expression following one assay but were not able to replicate this result, so we cannot confirm expression in amygdala.
Conclusion: We confirm expression of ΔFosB in the nucleus accumbens as seen in animal models exposed to opioids.
Current and Future Studies: We are further characterizing expression of ΔFosB in the amygdala, hippocampus, caudate, putamen, and other regions of the mesolimbic dopamine system. Moreover, we are concurrently working on isolating RNA for quantitative PCR.

Disclosures: L. Jabbour: None. **F. Cartee:** None. **M. Caisse:** None. **K. Haynes:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.04/SS58

Topic: G.08. Drugs of Abuse and Addiction

Support: UHCL Faculty Research Support fund

Title: A subtype-selective neuropeptide FF receptor antagonist attenuates morphine withdrawal syndrome

Authors: *D. H. MALIN, J. ELAYOUBI, M. M. HENCEROTH, J. R. CAMPBELL, C. MADISON, C. P. WARD
Human Sciences,, Univ. of Houston Clear Lake, Houston, TX

Abstract: Considerable evidence suggests the neuropeptide FF (NPFF) and related peptides exert anti-opiate actions, particularly at the supra-spinal level. Ventricular administration of NPFF antagonists or antibodies attenuates morphine tolerance and dependence, suggesting that the NPFF family contributes to these disorders. On the other hand, NPFF appears to exert pro-

opiate activity, particularly at the spinal level. Lameh *et al*, JPET 334: 244, 2010, suggested that activating the FF2 NPFF receptor subtype is responsible for pro-opiate effects, while activating the FF1 receptor subtype is responsible for anti-opiate effects. They also showed that the compound AC262620 selectively inhibits the FF1 receptor subtype. This experiment tested the hypothesis that the FF1 receptor is a major contributor to opiate dependence and subsequent withdrawal syndrome. The subjects were 18 male Sprague-Dawley rats. Osmotic minipumps infused each rat for seven days with morphine sulfate at a constant rate of 0.6mg/kg/hr. On the seventh day of infusion, morphine infusion was terminated by pump removal. After 23 hours, rats were injected i.p. with 10mg/kg of AC262620 (generously donated by Acadia Pharmaceuticals) or with saline/DMSO injection vehicle alone. Beginning forty-five minutes later, subjects were observed under blind conditions for 20 minutes on a standard checklist of somatically expressed morphine withdrawal signs. Student's t-test revealed a highly significant reduction in total morphine withdrawal signs from the vehicle control group to the AC262620 group, $t(16)=3.798$, $p=.001$. Differences in individual categories of signs were also significant in shakes, $t(16)=2.63$, $p=.009$ and in tremors, $t(16)=2.67$, $p=.0085$. Reductions in instances of writhes and vacuous chewing also approached significance, $p=.083$ and .093, respectively. These results are consistent with an earlier experiment where AC262620 administered to morphine-dependent rats reduced subsequent naloxone-precipitated withdrawal after a much lower rate of morphine infusion. Taken together, these findings support the hypothesis that NPFF or related neuropeptides contribute to opiate dependence and withdrawal syndrome through the FF1 receptor.

Disclosures: D.H. Malin: None. **J. Elayoubi:** None. **M.M. Henceroth:** None. **J.R. Campbell:** None. **C. Madison:** None. **C.P. Ward:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.05/SS59

Topic: G.08. Drugs of Abuse and Addiction

Support: PESCA Grant Program, Vice President for Research (VPR), Division of Research, Texas A&M University

Texas A&M Genomics Seed Grant; Texas A&M University Vice President for Research, Health Science Center, and AgriLife Research

Title: Exploring mechanisms for the effect of social environment on morphine response

Authors: *S. BATES[1], M. A. EMERY[2], C. T. HORRAX[5], P. J. WELLMAN[3], S. EITAN[4]
[2]Psychology, [1]Texas A&M Univ., College Station, TX; [3]Texas A&M Univ., College Sta, TX;

[4]Dept of Psychology, Behavioral & Cell. Neurosci Program, Texas A&M Univ., College Station, TX; [5]Neurosci., Texas A&M, College Station, TX

Abstract: Drug abuse is strongly influenced by socio-environmental factors. Our previous studies demonstrated that social housing conditions alter the propensity to acquire morphine reward and dependence in adolescent mice. Morphine-treated animals that are housed only with other morphine-treated animals (referred to as 'morphine only' animals) acquire morphine CPP significantly faster than morphine-treated animals which are housed with drug-naïve mice (referred to as 'morphine cage-mates'). Similarly, morphine only animals extinguished their morphine place preference at a markedly slower rate than the morphine cage-mates. This indicates a stronger and more robust acquisition of morphine CPP in animals housed with only other morphine-treated animals than in animals housed with drug-naïve animals. Lastly, morphine only animals display distinctly greater morphine withdrawal symptoms than morphine cage-mates. These results demonstrate that being housed with drug-naïve animals has a protective effect on development of opioid dependence, as well as the acquisition and maintenance of opioid reward. This further supports the notion that social conditions alter the propensity for developing opioid addiction.

Despite the preponderance of behavioral evidence, little is known about the mechanisms mediating the differential effects of social housing. In the current study, we explored possible mechanisms for the role of social environment on morphine response. In order to investigate this, we conducted qRT-PCR on gene targets that were identified using RNA-Seq. We also utilized DREADD technology to manipulate a novel DRG neuron subtype that was shown to be activated during social grooming.

We found that several genes were differentially altered between animals that were treated with the same drug, but that were housed differently (i.e., animals that were treated with saline or morphine and housed in 'only' conditions, or in mixed cages). These genes are from a variety of families and have diverse functions. Moreover, we identified a novel DRG neuron subtype that, when inhibited, decreases the protective effect of being housed with drug-naïve animals. Here, we report potential mechanisms for our social housing effect. In the future, more work should be done involving the targets identified here to develop a richer understanding of the role the social environment plays in morphine response. It is our hope that this will lead to better, more successful treatments for opiate use.

Disclosures: S. Bates: None. **M.A. Emery:** None. **C.T. Horrax:** None. **P.J. Wellman:** A. Employment/Salary (full or part-time):; Texas A&M University. **S. Eitan:** A. Employment/Salary (full or part-time):; Texas A&M University.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.06/SS60

Topic: G.08. Drugs of Abuse and Addiction

Support: T32 NS044928-13, NINDS

PhRMA Foundation Research Starter Grant

R01 DA039895-01A1, NIDA

Title: Changes in ventral tegmental area serum- and glucocorticoid-inducible kinase 1 (SGK1) catalytic activity and phosphorylation alter drug-related behaviors

Authors: *M. A. DOYLE[1], V. BALI[2], S. KASKA[3], S. E. COOPER[1], M. S. MAZEI-ROBISON[2]
[1]Neurosci. Program, [2]Dept. of Physiol., [3]Dept. of Pharmacol. and Toxicology, Michigan State Univ., East Lansing, MI

Abstract: Drugs of abuse are known to regulate activity of the mesolimbic dopamine system. Specifically, drug-induced changes in ventral tegmental area (VTA) cellular activity and gene regulation contribute to behavioral outputs associated with addiction. Our previous work has shown that serum- and glucocorticoid-inducible kinase 1 (SGK1) phosphorylation and catalytic activity are increased by chronic, but not acute, administration of cocaine and morphine. However, the functional significance of these changes in drug-related behaviors remains unclear. Thus, we have created novel viral constructs to overexpress SGK1 mutants in the VTA of adult mice, and I am assessing the impact of altered VTA SGK1 activity on drug reward via cocaine conditioned place preference (CPP) and voluntary morphine intake using a two-bottle choice task. Intra-VTA infusion of a catalytically inactive SGK1 mutant (K127Q) significantly decreased morphine intake and there was a similar non-significant trend for decreased cocaine CPP, suggesting that decreasing VTA SGK1 activity is sufficient to decrease drug reward and intake. I am now extending these studies by investigating the role of SGK1 Ser78 in drug-related behaviors, as phosphorylation of this site is increased by drugs of abuse. Intra-VTA infusion of SGK1 S78A, a mutant that prevents phosphorylation at S78, significantly decreased cocaine CPP as well as morphine intake and preference. Conversely, mice with VTA overexpression of SGK1 S78D, a mutant that mimics phosphorylation, exhibited a trend for increased morphine intake and preference. These data suggest that alteration of VTA SGK1 S78 phosphorylation is sufficient to modulate drug reward and/or intake. To more fully understand the role of VTA SGK1 in behaviors relevant to addiction, we have developed cre-dependent constructs to virally overexpress SGK1 mutants in a cell-type specific manner to determine whether SGK1 activity in DA or GABA neurons drives the observed behavioral effects. These studies, along with experiments utilizing a floxed-SGK1 mouse model, will allow identification of the specific cells and circuits that are critical for SGK1-mediated effects on drug behavior. Additionally, I have established a protocol for mouse cocaine self-administration and I am using this paradigm to determine whether alteration of VTA SGK1 activity is sufficient to modify operant drug intake or drug-seeking behavior, expanding our findings to more translationally-relevant models of addictive behavior. In summary, our goal is to characterize the role of VTA SGK1 activity in drug-related behaviors in order to assess SGK1's feasibility as a novel therapeutic target for addiction.

Disclosures: **M.A. Doyle:** None. **V. Bali:** None. **S. Kaska:** None. **S.E. Cooper:** None. **M.S. Mazei-Robison:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.07/SS61

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH R01 DA039895

NIH F31 DA042502

Title: Determination of circuit-specific morphological adaptations in ventral tegmental area dopamine neurons by chronic morphine

Authors: K. WHEELER[1], S. E. COOPER[2], *M. S. MAZEI-ROBISON[3]
[1]Neurosci., [2]Neurosci. Grad. Program, [3]Physiol., Michigan State Univ., East Lansing, MI

Abstract: Opiate drugs are the leading treatment for severe or chronic pain in the USA despite their extremely addictive properties. Chronic opiate exposure induces unique neuroadaptations in the mesocorticolimbic system, particularly in ventral tegmental area (VTA) dopamine (DA) neurons. For example, opiates reduce VTA DA neuron soma size, a change correlated with increased DA activity and reward tolerance. Prevention of this morphological change is sufficient to rescue the morphine-induced changes to behavior, suggesting its direct involvement in addiction-related processes. While VTA DA structural and functional plasticity are central to morphine reward and addiction, the molecular mechanisms driving these neuroadaptations remain elusive due to two main sources of VTA heterogeneity: multiple cell types and diversity within cell type based on projection target. To better understand the circuit-based consequences of morphine-induced neuroadaptations, we compared the morphology of VTA DA neurons that project to the nucleus accumbens (NAc) and medial prefrontal cortex (mPFC) in sham and morphine treated mice. We used stereotaxic bilateral infusion of retrograde Cre-dependent viral constructs (AAV2/5-DIO-eYFP and AAV5-DIO-mCherry) into DA neuron Cre-driver lines (tyrosine hydroxylase (TH)-Cre or dopamine transporter (DAT)-Cre). We found significant differences in basal soma size between established VTA subregions and therefore compared morphology within subregions for the remainder of the study. Chronic morphine treatment in TH-Cre mice significantly reduced the soma size of NAc medial shell-projecting neurons, but conversely had no effect on NAc lateral shell/core-projecting neurons. Interestingly, mPFC-projecting neuron soma size increased following chronic morphine treatment, despite the fact that the basal soma size of these neurons was larger than that of NAc-projecting neurons in sham-treated mice. We are now examining VTA DA neurons in DAT-Cre mice for validation of

morphine effects across DA-specific mouse lines commonly used in addiction research. Thus far, these data suggest that the unique morphological effect of chronic opiates on VTA DA morphology occurs within a distinct VTA DA population, and therefore direct manipulation of specific microcircuit/s may be critical to reverse the negative effects associated with chronic opiate use.

Disclosures: K. Wheeler: None. **S.E. Cooper:** None. **M.S. Mazei-Robison:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.08/SS62

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH ROI DA039895

Title: Determination of the upstream kinases responsible for increased phosphorylation of serum and glucocorticoid-inducible kinase by drugs of abuse

Authors: *V. BALI[1], M. A. DOYLE[2], S. E. COOPER[2], S. KASKA[3], M. S. MAZEI-ROBISON[1]
[1]Dept. of Physiol., [2]Neurosci. Program, [3]Pharmacol. and Toxicology, Michigan State Univ., East Lansing, MI

Abstract: Drugs of abuse alter the activity and output of dopamine neurons in the ventral tegmental area (VTA), which contributes to addiction-related behaviors. Our lab has previously shown that serum- and glucocorticoid-inducible kinase 1 (SGK1) phosphorylation and catalytic activity are increased by chronic administration of cocaine and morphine. Further, using viral-mediated overexpression studies, we have recently observed that decreasing VTA SGK1 catalytic activity decreases drug reward and intake, and that altering VTA SGK1 phosphorylation specifically at serine 78 (S78) is sufficient to modulate cocaine reward and morphine intake, as measured by conditioned place preference and a two-bottle choice task, respectively. However, little is known about regulation of SGK1 S78 phosphorylation, aside from its robust increase by drugs of abuse. Therefore, the aim of my study is to identify the kinases responsible for SGK1 S78 phosphorylation, as this may serve as a novel node for therapeutic intervention. Using *in silico* approaches, I identified promising candidate kinases, namely: glycogen synthase kinase 3 beta (GSK3β), cyclin-dependent kinase 5 (cdk5), extracellular signal-regulated kinase 5 (ERK5), and ERK1/2. As a first screen, I utilized the Neuro2A mouse neuroblastoma cell line and established an induction protocol that robustly increases both SGK1 catalytic activity and S78 phosphorylation. Briefly, I found that 24-hour serum starvation followed by re-feeding with complete media supplemented with epidermal growth factor or insulin for 1 hour significantly increased S78 SGK1 phosphorylation and

catalytic activity, as measured by phosphorylation of N-myc downstream regulated gene (NDGR) protein, the only known SGK1-specific substrate. I then evaluated the role of candidate kinases using pharmacological inhibitors: roscovitine (cdk5), GSK3β inhibitor XXVII (GSK3β kinase), U0126 inhibitor (ERK1/2) and BIX02189 (ERK5). My preliminary data indicate that inhibition of either cdk5 or GSK3β can dose-dependently decrease SGK1 catalytic activity and S78 phosphorylation. I am now completing a similar study to determine the potential roles of ERK1/2 and ERK5. Importantly, all of the inhibitors used in this study have been validated *in vivo*, so I will next test whether intra-VTA infusion is sufficient to prevent the cocaine- and morphine-induced increase in SGK1 S78 phosphorylation, as well as associated behavioral changes. Together, my work seeks to characterize the regulation of SGK1 S78 phosphorylation in drug-related behaviors to identify a potential novel pharmacological target for improved treatment of addiction.

Disclosures: V. Bali: None. **M.A. Doyle:** None. **S.E. Cooper:** None. **S. Kaska:** None. **M.S. Mazei-Robison:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.09/SS63

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH (NIDA) F31 DA042502

NIH (NIDA) R01 DA037426

Title: Determination of the morphine-induced transcriptome in ventral tegmental area dopamine neurons

Authors: *S. E. COOPER[1], K. WHEELER[2], M. MAZEI-ROBISON[3]
[1]Neurosci. Grad. Program, [2]Neurosci. Program, [3]Dept. Physiol., Michigan State Univ., East Lansing, MI

Abstract: Opiate exposure induces neuroadaptations in the mesocorticolimbic system, and in particular in ventral tegmental area (VTA) dopamine (DA) neurons. In rodents, chronic morphine reduced VTA dopamine (DA) neuron soma size, which directly correlated with increased DA activity and decreased reward, and prevention of this morphological change was sufficient to rescue the morphine-induced physiological and behavioral changes. While VTA DA structural and functional plasticity are central to morphine reward and addiction, the molecular responses driving these neuroadaptations remain elusive. To date, studies of drug-induced changes in VTA gene expression have been limited to the homogenization of the entire VTA,

which includes GABAergic and dopaminergic (tyrosine hydroxylase (TH)-positive) neurons. While several candidate genes have been identified with this approach, it is unknown whether these changes occur specifically in DA neurons and contribute to the structural neuroadaptations. To determine morphine-induced gene expression changes specifically in VTA DA or GABA neurons, we utilized Translating Ribosome Affinity Purification (TRAP). We crossed DA- (TH- or dopamine transporter (DAT)-Cre) and GABA-specific (vesicular GABA transporter (vGAT)-Cre) driver lines with Rosa26 EGFP-L10a mice, thereby allowing isolation of mRNA from VTA DA (THEGFP-L10a and DATEGFP-L10a) and GABA (vGATEGFP-L10a) neurons. In both DA-specific lines, we found significant enrichment of TH and DAT mRNA and depletion of GABAergic markers glutamic acid decarboxylase (GAD) and vGAT in bound fractions compared to input controls, consistent with successful purification. We first examined morphine-induced changes in the expression of candidate genes such as serum and glucocorticoid-regulated kinase 1 (SGK1). While SGK1 was induced by morphine treatment in the input control, consistent with whole VTA expression studies, this change was not detected in the bound, DA-specific, fraction. Thus, we hypothesize that the morphine-induced increase in SGK1 expression is driven specifically by VTA GABA neurons, which we will test using vGATEGFP-L10a mice. Given evidence that morphine-induced changes can be limited to specific cell types, we are now completing RNA sequencing analysis on samples from THEGFP-L10a and DATEGFP-L10a as an unbiased approach to identify changes that occur specifically in VTA DA cells. In this way, we hope to identify novel mechanisms that underlie opiate-induced neuroadaptations in the VTA in order to develop innovative targets for improved therapeutics.

Disclosures: S.E. Cooper: None. **K. Wheeler:** None. **M. Mazei-Robison:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.10/SS64

Topic: G.08. Drugs of Abuse and Addiction

Support: NIDA Intramural Funds

Ministerio de Economia y Competitividad SAF2014-54840-R

Fundacio La Marato de TV3 20140610

Government of Catalonia 2014-SGR-1236

Title: Functional μ-opioid- galanin Gal_1 receptor heteromers in the ventral tegmental area as targets for opioid use disorders

Authors: *N. CAI[1], E. MORENO[2], C. R. QUIROZ[1], W. P. REA[1], A. SEYEDIAN[1], C. LLUÍS[2], E. CANELA[2], V. CASADÓ[2], S. FERRE[1]
[1]NIDA/NIH, Baltimore, MD; [2]Neurobiologia Mol. Departament de Bioquímica i Biomedicina Mol., Univ. de Barcelona, Barcelona, Spain

Abstract: We recently demonstrated that μ-opioid- galanin Gal_1 receptor heteromers localized in the ventral tegmental area (VTA) constitute a target for the treatment of opioid use disorders. These heteromers can underlie previously described antagonistic interactions between the opioid and galanin systems, by which galanin antagonizes μ-opioid receptor-mediated signaling. We first detected μ-opioid-Gal_1 receptor heteromerization in mammalian transfected cells and obtained a specific peptide that disrupted the heteromerization and a negative crosstalk, by which galanin counteracted MAPK activation induced by the endogenous opioid agonist endomorphin-1. The negative crosstalk therefore constituted a biochemical property of the μ-opioid-Gal_1 receptor heteromer, which could also be identified *in situ* in VTA slices and *in vivo* with microdialysis experiments. Thus, galanin completely counteracted somatodendritic dopamine release induced by the local infusion of endomorphin-1. Both *in situ* and *in vivo* galanin-opioid interactions were also selectively counteracted by application of the disruptive peptide, demonstrating their dependence on μ-opioid-Gal_1 receptor heteromerization. These results indicated that dopaminergic cell function in the VTA is modulated by a predominant population of μ-opioid receptors forming heteromers with Gal_1 receptors. Using transfected mammalian cells, we are now addressing two questions in relation to opioid addiction in the frame of μ-opioid-Gal_1 receptor heteromers. First, based on the proof of concept that receptor heteromerization can change the properties of orthosteric ligands binding to any of the two molecularly different receptor units, we are screening μ-opioid ligands, with the aim of finding receptor agonists with lower affinity or efficacy or receptor antagonists with higher affinity for the μ-opioid receptor when forming heteromers with Gal_1 receptor. Second, based on the proof of concept that products of polymorphic variants of receptor genes can lead to different biochemical properties of receptor heteromers, we are investigating if the product of the single-nucleotide polymorphism rs1799971 (A118G) of the μ-opioid receptor gene induces significant changes in the functional and pharmacological properties of the μ-opioid-Gal_1 receptor heteromer. This common polymorphism, which results in the substitution of an asparagine for an aspartate in a putative extracellular glycosylation site, has been repeatedly associated with increased vulnerability to opiate abuse, but the mechanisms behind the putative loss of function of the modified μ-opioid receptor remain still to be established.

Disclosures: **N. Cai:** None. **E. Moreno:** None. **C.R. Quiroz:** None. **W.P. Rea:** None. **A. Seyedian:** None. **C. Lluís:** None. **E. Canela:** None. **V. Casadó:** None. **S. Ferre:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.11/SS65

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH Grant R01DA037257

NIH Grant GM09545902

NIH Grant R01DA03725703S1

Title: Drebrin regulates opiate-induced behavioral and structural plasticity in the NAc

Authors: *J. A. MARTIN[1], C. T. WERNER[1], Z.-J. WANG[1], J. N. SIEMIAN[1], P. ZHONG[2], D. HAGARTY[3], R. VISWANATHAN[1], R. L. NEVE[4], J.-Z. LI[1], R. CHANDRA[5], M. LOBO[5], A. M. GANCARZ[3], Z. YAN[2], D. M. DIETZ[1]
[1]Dept. of Pharm and Tox; Res. Inst. on Addictions; Program in Neurosci., [2]Dept. of Physiol. and Biophysics, State Univ. of New York At Buffalo, Buffalo, NY; [3]Dept. of Psychology, California State University, Bakersfield, Bakersfield, CA; [4]Dept. of Brain and Cognitive Sci., MIT, Cambridge, MA; [5]Dept. of Anat. and Neurobio., Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: Opiate addiction, a chronic relapsing disease, places a large societal and financial burden on our country. In particular, the fundamental issue for the therapeutic treatment of opiate addiction is the persistence of drug craving and seeking, leading to high rates of relapse following drug abstinence. These behaviors are characterized by long-lasting behavioral and cellular plasticity in key regions of the mesolimbic dopamine system, such as changes to the structure of and dendritic spine density on medium spiny neurons in the nucleus accumbens (NAc). To date, the cellular and molecular mechanisms governing opiate-induced structural plasticity remain undetermined. Following heroin self-administration, we find that expression of the essential actin-binding protein drebrin is decreased in the NAc, an effect that is transcriptionally regulated through the binding of HDAC2 at the drebrin promoter. Using viral-mediated gene transfer, the overexpression of drebrin attenuated, while CRISPR-Cas9 deletion exacerbated, heroin-primed (but not sucrose-primed) relapse-like behaviors. In addition, drebrin overexpression produced a downward shift, while CRISPR-Cas9 produced an upward shift, in within-session dose-response curves, demonstrating that drebrin regulates the reinforcing properties of heroin. The restoration of drebrin levels following heroin self-administration reversed the heroin-induced decrease in spine density and reductions in AMPA and NMDA receptor conductances. Taken together, these data demonstrate an essential role for drebrin in mediating the molecular mechanisms underlying opiate-induced behavioral and structural

plasticity. An understanding of these cellular responses will help lead to therapeutic interventions to prevent relapse.

Disclosures: **J.A. Martin:** None. **C.T. Werner:** None. **Z. Wang:** None. **J.N. Siemian:** None. **P. Zhong:** None. **D. Hagarty:** None. **R. Viswanathan:** None. **R.L. Neve:** None. **J. Li:** None. **R. Chandra:** None. **M. Lobo:** None. **A.M. Gancarz:** None. **Z. Yan:** None. **D.M. Dietz:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.12/DP12/SS66 (Dynamic Poster)

Topic: G.08. Drugs of Abuse and Addiction

Title: Chronic oxycodone exposire alters brain function and structure multimodal MRI study

Authors: *S. IRIAH[1], J. QIAO[1], L. TIMMS[1], P. P. KULKARNI[2], P. P. KULKARNI[2], C. F. FERRIS[3]
[1]Psychology, Northeastern Univ., Boston, MA; [2]Psychology, Northeastern Univ. Dept. of Psychology, Boston, MA; [3]Psychology, Northeastern University, Ctr. for Translational NeuroImaging, Boston, MA

Abstract: Abuse of prescription drugs is a major epidemic in the United States, accounting for over 19,000 deaths in 2014. Massachusetts alone, accounted for 1,200 of these fatalities. The combination of imaging methods and animal models has allowed identification of circuits of addiction and novel biomarkers that could guide clinical intervention. The present studies measure changes in brain function and structure using multimodal imaging in male rats exposed to two weeks of twice daily (2.5 mg/kg/injection) treatments of oxycodone (OXY). The treatments were conducted in a two-chamber testing arena to establish condition place preference. At the end of two weeks, vehicle controls (n = 10) and OXY treated rats (n = 10) were imaged in a 7.0T Bruker Biospec scanner using diffusion weighted imaging (DWI), resting state functional connectivity (fcBOLD) and quantitative, ultra-short time to echo contrast enhanced imaging (QUTE-CE). All voxel based measures for each modality were registered to a rat MRI atlas with 172 segmented, annotated brain regions. DWI and quantitative anisotropy was used to follow neuroadaptation or changes in gray matter microarchitecture in the mesencephalic dopaminergic system, limbic cortex and hippocampal complex. Resting state fcBOLD showed altered connectivity between these different brain regions while QUTE-CE showed changes in capillary density across several brain areas involved in pain processing between vehicle controls and OXY treated animals.

Disclosures: **S. Iriah:** None. **J. Qiao:** None. **L. Timms:** None. **P.P. Kulkarni:** None. **P.P. Kulkarni:** None. **C.F. Ferris:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.13/TT1

Topic: G.08. Drugs of Abuse and Addiction

Support: Startup Funds OSU-CHS

Title: Getting out of the net: Chronic escalating doses of morphine induces morphological changes in perineuronal nets and axonal guidance in substantia nigra in adolescent female rats

Authors: *R. GAGLIA[1], P. GONZALES[2], G. S. LARMOUR[1], M. T. BARDO[3], D. B. VAZQUEZ-SANROMAN[1]
[1]Anat. and Cell Biol., Oklahoma State Univ. Ctr. For Hlth. Scienc, Tulsa, OK; [2]Biol., Tulsa Community Col., Tulsa, OK; [3]Dept Psychol, Univ. Kentucky, Lexington, KY

Abstract: Mechanisms underlying adolescent morphine withdrawal in females are not fully understood. Changes in structural neuroplasticity can occur in GABAergic parvalbumin (PRV) positive neurons, which are enveloped by structures of the extracellular matrix called perineuronal nets (PNNs). In the current study, rats received chronic escalating morphine doses during the adolescent period, followed by immunofluorescence measurements for PNN morphology and expression in orbitofrontal cortex (OFC) and substantia nigra (SN). Adolescent female Sprague Dawley rats (PND 34) were administered increasing doses of morphine (5 to 25 mg/kg, s.c.) every 12 hours, for 6 consecutive days. Control rats received saline. On day 6, naloxone (2 mg/kg, i.p.) was injected 2 hr after the last morphine administration and somatic signs of withdrawal were recorded. We found that adolescent morphine escalating dose treatment resulted in the development of morphine dependence as measured by increased withdrawal signs (defecation, digging, mastication and grooming). A one-way ANOVA revealed a significant increase in the perimeter of PNNs [$F(1,76)=11.85$, $p<0.001$], and a reduction on the number of PNNs [$F(1, 3) = 25.36$, $p < .05$] and PNN intensity in SN [$X^2(1)= 8.96$, $p<0.05$] in the morphine-naloxone group compared to saline controls. Moreover, the morphine-naloxone group displayed a significant reduction in the number of PNNs in OFC [$F(1, 3) = 25.36$, $p < .05$]. These results indicate that morphine dependence produced by escalating dose treatment, followed by precipitated withdrawal, remodels PNNs surrounding GABA interneurons in SN and its indirect connections to OFC, suggesting a new possible cellular target where morphine-induced neuroplasticity takes place.

Disclosures: **R. Gaglia:** None. **P. Gonzales:** None. **G.S. Larmour:** None. **M.T. Bardo:** None. **D.B. Vazquez-Sanroman:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.14/TT2

Topic: G.08. Drugs of Abuse and Addiction

Support: Start up Funds OSU-CHS

Title: Perineuronal nets and neuronal cilia abnormalities in insular cortex of adolescent female rats treated with chronic escalating doses of morphine

Authors: *D. B. VAZQUEZ SANROMAN[1], R. GAGLIA[2], P. GONZALES[3], G. S. LARMOUR[1], M. T. BARDO[4], N. F. WILSON[1]
[1]Anat. and Cell Biol., Oklahoma State Univ., Tulsa, OK; [2]Anat. and Cell Biol., Oklahoma State Univ. Ctr. For Hlth. Scienc, Tulsa, OK; [3]Biol., Tulsa Community Colleague, Tulsa, OK; [4]Dept Psychol, Univ. Kentucky, Lexington, KY

Abstract: A negative emotional state resulting from withdrawal stress is common in opioid users, yet few studies have examined the effects of morphine withdrawal on extracellular matrix elements and neuronal cilia, both critical organelles that facilitate interactions between the cell and its environment in response to stressful events. Since the insular cortex (IC) is a key brain structure involved in the modulation of negative emotions, this study investigated morphine dependence produced by chronic escalating doses, followed by precipitated withdrawal, on perineuronal nets (PNNs) and neuronal cilia length in the IC. Adolescent female Sprague Dawley rats (PND 34) were administered increasing doses of morphine (5 to 25 mg/kg, s.c.) every 12 hours, for 6 consecutive days. Control rats received saline. On day 6, naloxone (2 mg/kg, i.p.) was injected 2 hr after last morphine administration. Somatic signs of morphine withdrawal were recorded and brains extracted for immunofluorescence analysis. We found that morphine escalating dose treatment resulted in the development of morphine dependence as measured by increased withdrawal signs (defecation and chewing). A Mann-Whitney test indicated that number of PNNs was significantly lower for morphine-naloxone rats (Mdn=10) than for saline rats (Mdn=21), $U= 3.15$, $p<0.05$. Moreover, we found that morphine-naloxone treated rats showed a significant increase in parvalbumin expression [$X2(1)= 7.91$, $p<0.01$]. Also, a separate Mann-Whitney test revealed that neuronal cilia length was significantly increased for morphine-naloxone rats (Mdn=9) compared to saline rats (Mdn=5), $U= 9.15$, $p<0.05$. The current findings demonstrate that the negative emotional state induced by precipitated opioid withdrawal is associated with neuronal cilia elongation and PNN remodeling in IC, perhaps suggesting a novel cellular target for treating opiate dependence and withdrawal.

Disclosures: **D.B. Vazquez Sanroman:** None. **R. Gaglia:** None. **P. Gonzales:** None. **G.S. Larmour:** None. **M.T. Bardo:** None. **N.F. Wilson:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.15/TT3

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH Grant AT007222

Honors College Distinguished Professorship

Chico Hyperbaric Center (Chico, CA)

Title: Contribution of oxygen and hyperbaric pressure to hyperbaric oxygen (HBO_2) suppression of naloxone-precipitated withdrawal in morphine-dependent mice

Authors: *R. M. QUOCK[1], P. N. MAHARAJ[1], S. N. DEWALD[1], E. K. HEATH[1], A. L. BREWER[1], D. Y. SHIRACHI[2]
[1]Psychology, Washington State Univ., Pullman, WA; [2]Physiol. and Pharmacol., Univ. of the Pacific, Stockton, CA

Abstract: Treatment with hyperbaric oxygen (HBO_2) has been reported to moderate signs of opiate withdrawal in human subjects [Epifanova *et al*., *Anesteziol. Reanimatol.* (3):34-9, 1995]. We recently reported that both 30- and 60-min treatments with HBO_2 suppressed naloxone-precipitated withdrawal signs in morphine-dependent mice [Nicoara *et al*., *Brain Res*. 1648:434-7, 2016]. Work in our laboratory seeks to identify possible mechanisms for the biomedical effects of HBO_2. This follow-up study was conducted to investigate the relative individual contributions of oxygen and hyperbaric pressure to suppression of withdrawal signs by HBO_2 treatment. Male NIH Swiss mice received s.c. injections of morphine sulfate or saline twice a day for 4 days, the daily morphine dose increasing progressively from 50 mg/kg on day 1 to 125 mg/kg on day 4. On day 5, different groups of animals were treated with normobaric air (NBA), normobaric 100% oxygen (NBO_2), hyperbaric air (HBA) or hyperbaric 100% oxygen (HBO_2) for 30 min. Normobaric was set at 1.0 atmosphere absolute (ATA), while hyperbaric was set at 3.5 ATA. Ninety min after NBA, NBO_2, HBA or HBO_2 treatment, opioid withdrawal was precipitated by i.p. injection of 5.0 mg/kg naloxone, and withdrawal signs—withdrawal jumping, wet-dog shakes, forepaw tremors, rearing, and defecation—were videorecorded for 30 min and later counted. Results showed a main effect of oxygen on rears, a main effect of hyperbaric pressure on tremors, and main effects for both oxygen and hyperbaric pressure on fecal boli. No significant results were found for withdrawal jumping or wet-dog shakes. Based on these

findings, the suppressant effect of HBO_2 on opioid withdrawal appears to be attributed to an effect of either oxygen or hyperbaric pressure or both, depending upon the specific endpoint under investigation. This intriguing result should be further studied to identify the neurochemical effects of both oxygen and hyperbaric pressure separately and in combination.

Disclosures: R.M. Quock: None. **P.N. Maharaj:** None. **S.N. DeWald:** None. **E.K. Heath:** None. **A.L. Brewer:** None. **D.Y. Shirachi:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.16/TT4

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH Grant AT007222

State of Washington Initiative Measure No. 171

Honors College Distinguished Professorship

Chico Hyperbaric Center (Chico, CA)

Title: Thalidomide-induced suppression for naloxone-precipitated withdrawal signs in morphine-dependent mice

Authors: *A. BREWER, P. K. MAHARAJ, S. N. DEWALD, E. K. HEATH, R. M. QUOCK
Psychology, Washington State Univ., Pullman, WA

Abstract: Chronic morphine treatment has been shown to increase glial activation in a pro-inflammatory manner in the hippocampus, posterior cingulate cortex and spinal cord indicating that glia may be involved in some aspects of morphine tolerance and dependence [Song and Zhao, *Neurosci. Res.* 39:281-86, 2001]. Glial inflammatory response causes an increase in the release of proinflammatory cytokines. Levels of tumor necrosis factor-α (TNFα) are reportedly elevated in the periaqueductal gray (PAG) of animals undergoing opioid withdrawal; moreover, microinjection of TNFα into the PAG induces withdrawal-like symptoms [Hao *et al*., *Neuropsychopharmacology* 36:664-76, 2011], suggesting TNFα may underlie some aspects of opioid withdrawal. Thalidomide has been shown to be a putative inhibitor of TNFα production in cell culture [Deng *et al*., *J. Invest. Dermatol.* 121:1060-65, 2003] and in human patients [Bauditz *et al*., *Gut* 50:196-200, 2002]. In order to investigate the role of TNFα in opioid withdrawal, male NIH Swiss mice received s.c. injections of morphine sulfate or saline twice a day for 4 days, the daily morphine dose increasing progressively from 50 mg/kg on day 1 to 125 mg/kg on day 4. On day 5, opioid withdrawal was precipitated by i.p. injection of 5.0 mg/kg naloxone.

Sixty min prior to the naloxone injection, different groups of mice were treated with either 65 mg/kg thalidomide or 30% DMSO vehicle. Withdrawal was videorecorded and assessed over a 30-min period using 5 different behavioral endpoints: withdrawal jumping; wet dog shakes; forepaw tremors; rearing; and defecation. Results showed that naloxone caused an increase in frequency of forepaw tremors and withdrawal jumps compared to saline-treated controls. Pretreatment with thalidomide reduced naloxone-precipitated withdrawal jumping, rearing, tremors and defecation but not wet-dog shakes. These findings support the working hypothesis that thalidomide can reduce certain physical signs of withdrawal in animals undergoing precipitated withdrawal. Further research will investigate whether thalidomide-induced inhibition of TNFα production is causally related to thalidomide suppression of opioid withdrawal.

Disclosures: A. Brewer: None. **P.K. Maharaj:** None. **S.N. Dewald:** None. **E.K. Heath:** None. **R.M. Quock:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.17/TT5

Topic: G.08. Drugs of Abuse and Addiction

Support: R00 DA038706-03

Title: Bidirectional effects of opioid self-administration on medial prefrontal cortex $GABA_BR$-GIRK signaling

Authors: *E. M. ANDERSON, L. FRIEDRICH, D. GOMEZ, M. C. HEARING
Dept. of Biomed. Sci., Marquette Univ., Milwaukee, WI

Abstract: Drug addiction is marked by subtle yet pervasive disruptions in cognitive function that facilitate its development and contribute to a high rate of relapse in addicts, even after sustained periods of abstinence. The prefrontal cortex (PFC) is a critical substrate for many higher-order cognitive functions, including decision making, inhibitory behavior, and processing of reward-related information. Physiological and functional abnormalities in medial PFC (mPFC) neurotransmission produced by repeated drug exposure have long been touted as an underlying factor in the development and persistence of addicted behavior. Layer 5/6 pyramidal neurons in the mPFC provide a key source of glutamate to limbic structures that mediates response to natural reward and drugs of abuse. Unlike psychostimulant exposure on mPFC pyramidal neuron physiology, almost nothing is known regarding the effects of opioids on mPFC pyramidal neuron physiology and inhibitory signaling. The present study sought to examine the impact of intermediate (10-14 d) and more prolonged (40+ d) opioid self-administration on Layer V/VI

pyramidal neuron intrinsic excitability and $GABA_BR$-GIRK signaling within sub-regions of the mPFC. Whole-cell recordings in acute-isolated brain slices from adult C57BL/6 mice showed that somatodendritic currents mediated by $GABA_B$ and GIRK1 are diminished 10-14 d and 40-45 d following intermediate remifentanil (0.05 mg/kg/inf) self-administration in the prelimbic cortex (PrL). These reductions were paralleled by increased reductions in the current required to evoke an action potential (increased excitability) and increased spike firing activity. In contrast, preliminary data indicate that more prolonged self-administration enhances $GABA_B$-GIRK signaling and reduces excitability of pyramidal neurons in the PrL. These data indicate that bidirectional alterations in GIRK signaling may represent early neuroadaptation critical for promoting opioid-seeking behavior during early drug use as well as reductions in mPFC function often associated with more habitual drug use. To this end, future studies will determine the anatomical and efferent selectivity of these adaptations within the mPFC and examine whether GIRK channels represent a potential therapeutic target for disrupting opioid relapse.

Disclosures: E.M. Anderson: None. **L. Friedrich:** None. **D. Gomez:** None. **M.C. Hearing:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.18/TT6

Topic: G.08. Drugs of Abuse and Addiction

Support: R00DA038706-03

Title: Cell-type and pathway-specific plasticity in the NAc following opioid self-administration

Authors: A. MADAYAG[1], L. FRIEDRICH[2], *M. C. HEARING[3]
[1]Pharmacol., UNC Chapel Hill, Chapel Hill, NC; [3]Dept. of Neurosciences, [2]Marquette Univ., Milwaukee, WI

Abstract: Unlike psychostimulants, little is known of opiate-induced synaptic plasticity at excitatory synapses in the nucleus accumbens (NAc) and its relevance to drug-seeking. Medium spiny neurons (MSNs), the principal NAc cell-type, typically express either the dopamine receptor 1 (D1-MSNs) or dopamine receptor 2 (D2-MSNs), the presence of which determines cell physiology and contribution to drug-related behaviors. BAC transgenic mice expressing fluorescent proteins under the control of D1R or D2R promoters were used in combination with whole-cell electrophysiology recordings in acute brain slices to identify opiate-induced plasticity in NAc shell and core MSN glutamate signaling following 10-14 d and 45 d withdrawal from remifentanil self-administration. AMPAR/NMDAR (A/N) ratios and AMPAR-mediated miniature excitatory postsynaptic currents (mEPSCs) were increased in D1-MSNs of the NAc

shell, whereas mEPSCs were reduced in D2-MSNs following 10-14 d of home-cage abstinence. Preliminary data examining cell- and pathway-specific plasticity using ex vivo optogenetic stimulation and retrograde tracers indicates that infralimbic-to-NAc shell synaptic strength and transmitter release probability is enhanced at D1-MSNs that project to the ventral pallidum. Interestingly, unlike non-contingent opioid exposure, mEPSC signaling is also elevated in D1-MSNs of the NAc core, suggesting that using a model of drug administration that requires performance of an operant task engages opioid-induced plasticity within this region. These data identify novel cell-type and pathway-specific alterations in synaptic strength and transmission that are distinct and overlapping in locus and timeline from those produced by psychostimulants and non-contingent opioid exposure. Future experiments will examine potential mechanisms underlying the cell-type plasticity and determine the relevance of cell- and pathway-specific plasticity in relapse behavior.

Disclosures: **A. Madayag:** None. **L. Friedrich:** None. **M.C. Hearing:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.19/TT7

Topic: G.08. Drugs of Abuse and Addiction

Support: DA09082

Title: Opioid-induced adaptations in the neuropeptide Y system in the rat locus coeruleus

Authors: *C. C. THEISEN[1], B. A. S. REYES[2], E. J. VAN BOCKSTAELE[2]
[2]Pharmacol. & Physiol., [1]Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: Opioid addiction is a serious public health problem in the United States, with over 2.5 million people currently addicted to heroin or prescription opioids. Stress is implicated as a risk factor in vulnerability to opioid abuse and relapse. Recently, neuropeptide Y (NPY) has emerged as a neurochemical mediator of stress resilience. Treatment with NPY prevents and reverses hyperarousal, anxiety, and depression-like behavior rat models of post-traumatic stress disorder, while low levels of NPY are associated with stress vulnerability. In healthy subjects, NPY levels are positively correlated with feelings of dominance, confidence, and superior performance under stress. Interestingly, male rodents exhibit a 2-fold increase in plasma NPY following stress exposure compared to females, indicating NPY may be a mediator of sex differences in stress resilience. Such studies suggest NPY modulates stress resilience, and aberrant NPY activity may increase susceptibility to stress-related psychiatric illness, particularly in females. Here, we investigate adaptations in the endogenous NPY system following chronic morphine exposure and naltrexone-induced withdrawal in the LC of male and female Sprague Dawley rats. Rats were

implanted with placebo or slow release morphine pellets (2 x 75 mg) to induce morphine dependence. After 6 days, rats received i.p. saline or naltrexone (100 mg/kg) to induce acute withdrawal. Rats were euthanized 30 minutes post-injection, and brains were harvested and dissected. Western blot analysis indicated baseline NPY levels were comparable in males and females. Unpaired t-tests for placebo and morphine rats revealed chronic morphine exposure caused significant decreases in LC NPY expression in males ($P < 0.01$) and females ($P < 0.01$). However, ANOVA of male and female placebo, morphine, and withdrawal groups, showed sex differences in the effect of morphine exposure and withdrawal on NPY. A post-hoc Tukey's test found chronic morphine exposure led to a larger decrease in NPY levels in females, compared to morphine-treated males ($P < 0.01$). Additionally, in males, withdrawal from morphine caused a greater decrease in NPY levels, compared to chronic morphine exposure alone ($P < 0.05$). The decrease in NPY levels observed in males subjected to withdrawal was not significantly different from the decreased levels in females exposed to chronic morphine alone. These findings demonstrate significant and sexually dimorphic adaptations in NPY signaling following chronic opioid exposure and withdrawal, and have important implications for the development of therapeutics to target the NPY system and increase resilience and resistance to relapse.

Disclosures: C.C. Theisen: None. **B.A.S. Reyes:** None. **E.J. Van Bockstaele:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.20/TT8

Topic: G.08. Drugs of Abuse and Addiction

Support: NIH-NIAAA NRSA F31AA024033

NIH 1R01AA023797

Title: Modeling functional genetic variation at the mu opioid receptor using human stem cells

Authors: *A. HALIKERE[1], J. C. MOORE[2], M. SWERDEL[3], J. A. TISCHFIELD[2], R. P. HART[3], Z. PANG[1]
[1]Dept. of Neurosci. and Cell Biol., CHINJ, RWJMS, Rutgers Univ., New Brunswick, NJ;
[2]Genet., RUCDR/Infinite Biologics, The Human Genet. Inst. of New Jersey, Piscataway, NJ;
[3]Cell Biol. and Neurosci., The Human Genet. Inst. of New Jersey, Rutgers Univ., Piscataway, NJ

Abstract: Significant genetic variability in the mu opioid receptor (MOR) is thought to impact individual responses to opioid drugs. The most common single nucleotide polymorphism (SNP) rs1799971 in the MOR (OPRM1 A118G) replaces an asparagine with an aspartate at position 40 (MOR N40D) of MOR. Previous functional studies investigating the effect of N40D on the

signaling profile of the mu opioid receptor have been controversial and unclear, largely due to the lack of an effective system to model this SNP. To develop a more simplified and species-specific system to provide new insight into the molecular and cellular consequences of OPRM1 A118G, we used CRISPR/Cas9 gene editing to generate two sets of isogenic human stem cell lines carrying homozygous N40 or D40 alleles from which we generated homogeneous populations of human induced neuronal cells (iNs) for functional analyses. Isogenic D40 MOR expressing iNs consistently manifest a stronger suppression of inhibitory synaptic release than N40 MOR human neuronal cells, suggesting acute signaling may be enhanced. Moreover, our data also suggest that MOR N40D variants may regulate MOR function differentially in long term exposures. Specifically, N40 MOR iN cells regained sensitivity to DAMGO following chronic (7 days) agonist exposure whereas D40 MOR iNs failed to show re-sensitization, suggesting potential differences in membrane recycling dynamics. Not only do isogenic cell lines exhibit consistent synaptic phenotypes, but patient derived induced pluripotent stem cell-derived iN cells exhibit similar cellular and synaptic differences in MOR regulation as well, providing strong evidence to support these new insights on MOR function. Our data illustrates the strength of using human pluripotent stem cells to isolate cellular and molecular mechanisms underlying human genetic variation.

Disclosures: A. Halikere: None. **J.C. Moore:** None. **M. Swerdel:** None. **J.A. Tischfield:** None. **R.P. Hart:** None. **Z. Pang:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.21/TT9

Topic: G.08. Drugs of Abuse and Addiction

Title: Examinations of morphine-induced withdrawal and its related comorbidity

Authors: *C.-Y. OU[1], C.-W. WU[2], A. HUANG[3]
[1]Dept. of Psychology, Fo Guang Univ., Jiaosi, Taiwan; [2]Dept. of Pharm., Keelung Hospital, Ministry of Hlth. and Welfare, Keelung City, Taiwan; [3]Fo Guang Univ., Yilan County, Taiwan

Abstract: Establishing morphine induces withdrawal symptoms and its related comorbidity such as anxiety behavior is a crucial to understand the brain mechanism of drug addiction. This present study addresses this issue. In the beginning of this experiment, all of rats were given a morphine-withdrawal procedure. During this period of time, rats were intraperitoneally injected a specific dose of morphine in the morning and then injected another different dose of morphine in the afternoon as following. Respectively, on Day1, they were given 5 mg/kg morphine and 10 mg/kg morphine. On Day 2, rats were given 15 mg/kg and 20 mg/kg morphine. On Day 3, rats were injected 25 mg/kg and 30 mg/kg morphine. On Day 4, rats were given 35 mg/kg and 40

mg/kg. On Day 5, all rats were given 50 mg/kg morphine and three hours later, all rats were given 2mg/kg naloxone to form withdrawal effects. After 15 minutes, rats were randomly tested anxiety behaviors and motor function in the zero maze and in the open field test. The results indicated that morphine group can induce withdrawal sign in the higher lose weights and stronger anxiety behaviors in lower crossing numbers and lower spent time in the central square. However, non-significant differences for spent time in open arm and staring latency time between morphine and control groups, indicting morphine group has no anxiety behaviors occurred in the zero mazes. In the open field test, morphine group showed a lower total distance travelled than control group, indicating morphine withdrawal effect might reduce motor function during the withdrawal phase. The present findings might offer some insights for interventions in morphine's withdrawal symptoms and its comorbidities. These data should be discussed further.
Keywords: morphine, withdrawal, anxiety, motor function, rat

Disclosures: **C. Ou:** None. **C. Wu:** None. **A. Huang:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.22/TT10

Topic: E.03. Basal Ganglia

Support: CNRS

University of Strasbourg

ANR

NARSAD

Title: Reconsidering the definition of the tVTA (tail of the ventral tegmental area) or RMTg (rostromedial tegmental nucleus)

Authors: *F. FAIVRE[1], C. FILLINGER[1], D. DANIEL[1], J. KAUFLING[2], E. DESCHATRETTES[3], D. PLASSARD[4], P. ROMIEU[3], J. SWILLER[3], P.-E. LUTZ[1], C. SANDU[1], C. THIBAULT-CARPENTIER[4], D. MASSOTTE[1], P. VEINANTE[1], M. BARROT[1]
[1]CNRS UPR 3212, STRASBOURG CEDEX 03, France; [2]MRC Brain Network Dynamics Unit, Univ. of Oxford, Oxford, United Kingdom; [3]LNCA UMR7364, Strasbourg, France; [4]IGBMC UMR7104, Illkirch CEDEX, France

Abstract: In the past decade, a new mesopontine structure was described in the rat brain: the tail of the ventral tegmental area (tVTA) or rostromedial tegmental nucleus (RMTg). This brain region attracted a lot of attention as it densely innervates the VTA and substantia nigra,

providing the most potent inhibitory GABA control of dopamine systems. It was also proved to be the anatomical target by which opiates recruit dopamine systems. The tVTA is the main output of the lateral habenula, and its relay toward dopamine systems. It controls avoidance behaviors and aversive responses, encodes reward prediction errors··· However, this structure is not yet present in rat and mouse brain atlases, and some questions regarding the definition and boundaries of the tVTA remain. The aim of our study was to reconsider and compare these definitions (neurochemical, anatomical, functional and genetic) in the rat brain. First, we did a side-by-side comparison of the various parameters defining the tVTA, by doing immunohistochemistries based on different protein markers of the tVTA: μ opioid receptor, glutamate decarboxylase 67, NeuN, and c-Fos and FosB/ΔFosB after acute administration or self-administration of cocaine. The results allowed us providing a stereological analysis and a 3D reconstruction of the structure. In a second step, we verified, by tract-tracing, the lateral habenula-tVTA-VTA connections. The results show that: 1) the various neurochemical markers of the tVTA define a bilateral structure with same localization and boundaries within the midbrain; 2) the tVTA contains about 2500 neurons per side and most of them can be recruited by cocaine (~60%); 3) the neurochemistry-based and connectivity-based definitions are not matching perfectly, and the neurochemical definition seems to be more relevant.

Disclosures: F. Faivre: None. **C. Fillinger:** None. **D. Daniel:** None. **J. Kaufling:** None. **E. Deschatrettes:** None. **D. Plassard:** None. **P. Romieu:** None. **J. Swiller:** None. **P. Lutz:** None. **C. Sandu:** None. **C. Thibault-Carpentier:** None. **D. Massotte:** None. **P. Veinante:** None. **M. Barrot:** None.

Poster

422. Opioid Cellular Physiology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 422.23/TT11

Topic: E.03. Basal Ganglia

Support: CNRS

University of Strasbourg

Title: The tail of the ventral tegmental area: aversive drugs, painful stimuli, stressful stimuli, opioid withdrawal and conditioned taste aversion

Authors: *M. BARROT[1], F. FAIVRE[1], I. YALCIN[1], M.-A. MULLER[2], D. MASSOTTE[1], M. MAJCHRZAK[2], M.-J. SANCHEZ-CATALAN[3]
[1]INCI - CNRS UPR3212, Strasbourg, France; [2]LNCA UMR7364, Strasbourg, France; [3]Dept. de Farmacia y Tecnología Farmacéutica y Parasitología, Universitad de Valencia, Burjassot (Valencia), Spain

Abstract: The tVTA (tail of the ventral tegmental area) or RMTg (rostromedial tegmental nucleus), is a GABA mesopontine structure controlling dopamine neurons. It has been implicated in the control of aversive responses, and the stimulation of the LHb/tVTA pathway favors avoidance behaviors. These data suggested an important role of the tVTA in the response to aversive situation. The goal of our study was first to test the influence of various aversive stimuli on the Fos induction in the tVTA in rats and, in a second time, to evaluate the behavioral consequences of a tVTA lesion on opiate withdrawal and on lipopolysaccharide (LPS) and lithium chloride (LiCl) induced conditioned taste aversion (CTA). We tested different types of aversive stimuli, including electric foot shock, pain, predator odor... and we used the response to amphetamine as positive control to compare the Fos responses. We show that only repeated foot-shocks (but not single ones) induced Fos in the tVTA. Naloxone-precipitated opiate withdrawal was also able to induce Fos in mu-opioid receptor positive and negative tVTA cells, which is supportive of both direct and indirect tVTA recruitment. Finally, a bilateral lesion of the tVTA had no impact on CTA and on the behavioural signs of precipitated opiate withdrawal. In conclusion, while stimulation of the tVTA favors avoidance behaviors, our data show that the processing of aversive information may not be a characteristic of the tVTA and that the tVTA does not necessarily process behavioural responses to all aversive stimuli.

Disclosures: M. Barrot: None. **F. Faivre:** None. **I. Yalcin:** None. **M. Muller:** None. **D. Massotte:** None. **M. Majchrzak:** None. **M. Sanchez-Catalan:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.01/TT12

Topic: H.01. Animal Cognition and Behavior

Title: Serotoninergic projections to the OFC and BLA modulate reversal learning

Authors: *D. TAPP, M. MCMURRAY
Psychology, Miami Univ., Oxford, OH

Abstract: Behavioral flexibility, the ability to adapt to changing reward contingencies, is a critical aspect of choice behavior. Such ability is disrupted in numerous psychiatric disorders, such as substance abuse disorders, attention deficit disorder, and obsessive-compulsive disorder. The orbitofrontal cortex (OFC) and the basolateral amygdala (BLA) have been implicated as key regulators for this behavior. Additionally, the neurotransmitter serotonin is known to influence behavioral flexibility, and is disrupted in numerous psychiatric disorders. While serotonin and these brain regions have been examined separately, they have yet to be directly linked in this behavioral context. Using a rat model, this study examined such a relationship by selectively leisoning serotoninergic projections to the OFC, BLA, or both regions with a SERT-conjugated

Saporin, and assessing behavioral flexibility in a probabilistic spatial reversal-learning task. Preliminary results indicated that the loss of serotonergic projections to either the OFC, BLA, or both impaired behavioral flexibility. Based on these results, we determined that serotonin regulates reversal learning through its action in the OFC and BLA. Therefore, the serotonergic system may serve as a future therapeutic target for diseases in which behavioral flexibility is impaired, and may explain the effectiveness of serotonin modulators in the treatment of these diseases.

Disclosures: D. Tapp: None. **M. McMurray:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.02/TT13

Topic: H.01. Animal Cognition and Behavior

Support: KAKENHI 26290009

KAKENHI 14320

Brain/MINDS

Title: Action-specific reinforcement and update by direct- and indirect- pathway striatal projection neurons

Authors: *S. NONOMURA[1], K. NISHIZAWA[2], K. KOBAYASHI[3], Y. SAKAI[1], Y. KAWAGUCHI[4], A. NAMBU[4], K. YAMANAKA[5], K. ENOMOTO[1], Y. ISOMURA[1], M. KIMURA[1]
[1]Tamagawa Univ., Machida / Tokyo, Japan; [2]Dept Mol Genet, Fukushima Med. Univ., Fukushima, Japan; [3]Fukushima Med. Univ., Fukushima 960-1295, Japan; [4]Natl. Inst. Physiol Sci., Okazaki, Japan; [5]Physiol., Juntendo University, Hlth. and Sports Sci., Chiba, Japan

Abstract: In order to reach a goal, we take an optimal action by flexibly selecting it from a large number of repertoire based on learning from positive and negative outcomes of our decision. On this occasion, proper assignment of reinforcement to specific decision and action is critical but it is generally difficult. Although the basal ganglia are thought to play major roles in these processes, neuronal basis remains largely unclear. We addressed this issue by focusing on the roles of direct and indirect pathways. We used transgenic rats (Long Evans) expressing Cre recombinase under the promotor of tachykinin receptor 1 (TAC1-Cre rat, for direct pathway) or dopamine D2 receptor (Drd2-Cre rat, for indirect pathway). We injected adeno-associated virus (AAV) vectors that express channelrhodopsin2 (ChR2) in a Cre-dependent manner unilaterally into the dorsomedial striatum (DMS). A 32-ch silicon probe was inserted vertically into the

DMS, and an optic fiber was introduced obliquely to illuminate LED light (460 nm for ChR2, < 2 mW) at the recording sites. Neurons were identified as direct- (dSPNs) or indirect- (iSPNs) pathway striatal projection neurons based on the excitatory response evoked by LED light. Rats performed a free choice task for probabilistic reward under head-fixed condition: they held a lever with right forearm at the center position, pushed or pulled it after visual GO signal, and kept it in the target zones for more than 300 ms to wait for reward (R+) or no-reward (R–) tones and actual outcomes. PUSH and PULL were assigned as either high-value (reward at 80% of trials) or low-value (20%) for 40-60 trials, then action-outcome contingency was reversed. We identified 52 dSPNs from 4 rats and 42 iSPNs from 5 rats. We observed (1) Most iSPNs (37/42) were activated by (R–), whereas most dSPNs (42/52) were activated by (R+) ; (2) Responses of iSPNs to (R–) and those of dSPNs to (R+) occurred preferentially after either PUSH or PULL (32/37, 87% of iSPNs and 32/42, 76% of dSPNs) and the other neurons exhibited similar responses after PUSH and PULL; (3) (R–) responses of iSPNs with preferred action were larger when it was followed by switch choices in the next trial than by stayed choices (22/32) whereas these responses with non-preferred action did not change according to subsequent switch or stayed choses. Our results indicate that direct and indirect pathways of the basal ganglia reinforce and update chosen actions in differential manner. They provide a new insight into neuronal mechanisms of action-specific reinforcement and selection in the basal ganglia, and implications for understanding neuronal basis of basal ganglia disorders.

Disclosures: S. Nonomura: None. **K. Nishizawa:** None. **K. Kobayashi:** None. **Y. Sakai:** None. **Y. Kawaguchi:** None. **A. Nambu:** None. **K. Yamanaka:** None. **K. Enomoto:** None. **Y. Isomura:** None. **M. Kimura:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.03/TT14

Topic: H.01. Animal Cognition and Behavior

Support: NIH grant R01-MH104494

Title: Neuronal activity in the primate amygdala during economic decisions

Authors: *A. JEZZINI[1], C. PADOA-SCHIOPPA[2]
[1]Washington Univ. In St Louis, Saint Louis, MO; [2]Washington Univ. in St Louis, Saint Louis, MO

Abstract: Experimental and theoretical results of recent years suggest that different groups of neurons in the orbitofrontal cortex (OFC) participate in the formation of good-based economic decisions. In contrast, relatively less is known about the contribution of other areas to this mental

process. A primary candidate is the amygdala, which, together with OFC, is the only brain area where lesions affect performance in reinforcer devaluation tasks. Other research groups found value-coding cells in this area (Paton et al 2006; Grabenhorst et al 2012). However, differences in experimental design and data analysis prevent direct comparisons with our results. In this study, we recorded from the amygdala of rhesus monkeys engaged in a juice choice task.
The experimental design was as in studies of OFC (Padoa-Schioppa and Assad, 2006). In each session one animal chose between two juices offered in variable amounts. Juice quantities varied from trial to trial, and the animal indicated its choice with a saccade. Choice patterns presented a quality-quantity trade-off, and the subjective relative value of the two juices was derived from a sigmoid fit on a session-by-session basis.
We recorded and analyzed the activity of 397 neurons. Firing rates were examined in several time windows. A "trial type" was defined by two offers and one choice (e.g., [1A:3B, A]). A "neuronal response" was defined as the activity of one cell, in one time window, as a function of the trial type. Task-related responses were identified with an ANOVA ($p<0.001$) and regressed against a large number of variables potentially encoded in this region. These included offer value, chosen value, other value, total value, etc. (19 total).
Two procedures for variable selection (stepwise and best-subset) identified three variables that best explained the whole population: offer value, chosen value and chosen juice. Remarkably, these are the same variables previously identified in OFC. The most notable difference between the two areas appeared to be the prevalence of different cell groups. Specifically, chosen value cells were much more prevalent in the amygdala, while offer value cells were much more prevalent in the OFC.

Disclosures: **A. Jezzini:** None. **C. Padoa-Schioppa:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.04/TT15

Topic: H.01. Animal Cognition and Behavior

Support: KAKENHI 17022052

KAKENHI 22300138

KAKENHI 25282246

KAKENHI 16H03301

KAKENHI 15J00709

Title: The effect of serotonin 5-HT_4 receptor antagonist on discounting of reward value in decision making

Authors: F. AKIZAWA[1], T. MIZUHIKI[2], R. KUBOKI[1], *M. SHIDARA[2]
[1]Grad Sch. of Comprehensive Human Sci., Univ. of Tsukuba, Tsukuba-shi, Ibaraki, Japan;
[2]Univ. Tsukuba, Tsukuba-shi/Ibaraki, Japan

Abstract: When we choose an action from some alternatives in order to acquire reward, we usually compare each reward value that depends on the balance between reward amount and the cost to obtain it. Previous studies have shown that animals and humans tend to choose smaller immediate reward than larger delayed reward when the serotonin (5-HT) level in the brain decreases, suggesting that serotonin modulates the temporal discount factor of the reward value when getting reward after waiting. On the other hand, it is said that basal ganglia-thalamocortical loop is involved in the reward-based decision-making and 5-HT might modulate the function of some of the loop structures. Recently, Hori et al. (2016, the 39th Annual Meeting of the Japan Neuroscience Society) reported that 5-HT4 receptor antagonist increased the temporal discount factor of reward value in the reward delay task. Since 5-HT4 receptors are largely distributed in striatum of macaque monkeys, this suggests the role of 5-HT4 receptor in the basal ganglia on the temporal discount factor in the reward-based decision-making. Here, we used a decision-making schedule task to assess the role of 5-HT4 receptor on choices depending on reward value. In the task, a monkey was required to choose one of two alternatives. Two kinds of choice cue presented side by side on the monitor were randomly picked up from 16 different combination of reward amount and schedule: 1, 2, 3 or 4 drops of liquid reward by 1, 2, 3 or 4 repeats of a bar-release trial. Brightness and length of the choice target indicates the amount of reward and the number of trial, respectively. Following a choice of one target by touching either left or right bar in front of the monkey, the chosen reward schedule task (consisted of a bar-release trial detecting the target color change from red to green) was started. After the monkey learned this task, we administrated 5-HT4 receptor antagonist GR125487 (1 mg/kg a day) or vehicle systemically and compared the discount factor between two conditions. The value of discount factor k was estimated by fitting the monkey's choice probabilities using exponential discounting model of reward value. Preliminary data from one monkey showed that the discount factor k was higher in GR125487 administration than in control condition. Moreover, in GR125487 administration, the discounting of reward value in schedule task part measured by error rates was also larger than that in control condition. These results suggest that 5-HT4 receptor in basal ganglia may play a role in regulating the discounting of reward value in decision making processing.

Disclosures: **F. Akizawa:** None. **T. Mizuhiki:** None. **R. Kuboki:** None. **M. Shidara:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.05/TT16

Topic: H.01. Animal Cognition and Behavior

Support: JSPS KAKENHI [15H05917]

JSPS KAKENHI [15H06872]

AMED Strategic Research Program for Brain Sciences

Title: Encoding expected reward value for formulating goal-directed decision in the rostro-medial caudate and the ventral pallidum

Authors: *A. FUJIMOTO[1], Y. HORI[1], Y. NAGAI[1], K. W. MCCAIRN[2], T. HIRABAYASHI[1], E. KIKUCHI[1], M. TAKADA[2], T. SUHARA[1], T. MINAMIMOTO[1]
[1]Dept. of Functional Brain Imaging Res., Natl. Inst. of Radiological Sciences, QST, Chiba, Japan; [2]Primate Res. Inst., Kyoto Univ., Inuyama, Japan

Abstract: The limbic basal ganglia networks are conjectured to mediate neural activity underlying reward prediction and goal-directed behavior. Having recently demonstrated that the rostromedial caudate (rmCD) is essential for normal goal-directed decision based on reward size (Nagai et al., 2016), the ventral pallidum (VP)—an area that is reciprocally connected with the rmCD—has also been implicated in reward-based neural processing. How rmCD and VP interact, however, and formulate value-based decisions remain unclear. To address this, we recorded neuronal activity from the rmCD and the VP from rhesus monkeys (N=2), while they performed the 'reward-size' task (Minamimoto et al., 2009). In this task, the monkeys performed an instrumental action (lever release < 1 sec after a 'go' signal) following presentation of a cue associated with the reward size (1, 2, 4, or 8 drops of juice). Both rmCD (39/107) and VP (63/105) transiently encoded the expected reward size following a cue presentation. Neuronal latency for the value coding in the rmCD neurons was significantly longer than the VP neurons (rmCD: 235 ms, VP: 115 ms, $P < 0.01$), suggesting faster recruitment of the VP rather than the rmCD in the value expectation process. To examine the causal role of these two regions, we inactivated bilateral VP by local infusion of $GABA_A$ receptor agonist (muscimol) in one monkey, and compared the behavioral effects with those from rmCD inactivation (N=2, Nagai et al., 2016). Silencing in either structure impaired the normal relationship between error rate and reward size, while the VP inactivation resulted in higher error rates than the rmCD inactivation. These results imply that both the rmCD and the VP encode an expected reward signal, which is critical for formulating decisions in goal-directed behavior. In conclusion, the VP encodes reward and goal-directed behavior in a complex, reciprocating manner, with rmCD.

Disclosures: **A. Fujimoto:** None. **Y. Hori:** None. **Y. Nagai:** None. **K.W. McCairn:** None. **T. Hirabayashi:** None. **E. Kikuchi:** None. **M. Takada:** None. **T. Suhara:** None. **T. Minamimoto:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.06/TT17

Topic: H.01. Animal Cognition and Behavior

Support: Big Brain Theory program (BBT) - Brain and Spine Institute (ICM)

"For Women in Science" - Fondation L'Oréal

Title: Probing the motivational control of decision making in mice using an in-house automatized, high throuhput behavioral task

Authors: *C. SCHREIWEIS, M. GACOIN, G. PENDERIA, J. DAUNIZEAU, E. BURGUIÈRE
Brain and Spine Inst. (ICM), Paris, France

Abstract: Anticipated costs and benefits of cognitive control are predicted to be strategically balanced. The expected value of control (EVC), a measurement of worthiness of investing executive resources in a decision, has been suggested to be computed by the anterior cingulate cortex (ACC), a pathologically deviating activation of which is associated with disorders such as ADHD and OCD. Recent neurocomputational studies predict that choices are only executed once passing a critical decision threshold, which can be dynamically adjusted, involving the basal ganglia. Here, we study the motivational control of decision making in mice, using an ecologically designed decision making task, during which task difficulty and reward amounts vary. Subjects live in the operant chambers during the behavioural task, where every event is automated through an in-house software management of the task (e.g. stimulus presentation, food delivery, optogenetic stimulation). Preliminary data suggest that task performance negatively correlates with increasing difficulty levels and drops during the inactive time of day (daytime), strongly supporting the automatized, high-throughput, ecological task design. In a first version of the task, we additionally observed an interference of conflicting learning strategies: during early task performance, the acquisition of the quality of stimuli was overlaid by a win-stay/lose-shift strategy. In the future, we will apply in vivo electrophysiological recordings and optogenetic modulation to test our working hypothesis that costs of decision making is encoded by active, prefrontal cortical inhibition of premature selection in basal ganglia.

Disclosures: **C. Schreiweis:** None. **M. Gacoin:** None. **G. Penderia:** None. **J. Daunizeau:** None. **E. Burguière:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.07/TT18

Topic: H.01. Animal Cognition and Behavior

Support: ZIA MH002928

Title: Different time courses of explore-exploit decisions in primate amygdala, striatum, and orbitofrontal cortex

Authors: *V. D. COSTA, B. B. AVERBECK
Lab. of Neuropsychology, NIMH/NIH, Bethesda, MD

Abstract: Many decisions involve a tradeoff between exploiting existing knowledge of the world to obtain rewards right now, versus exploring something unknown to gain a potential advantage in obtaining future rewards. This is referred to as the explore-exploit dilemma. Managing this tradeoff is important in dynamic environments, where new options periodically become available or the values associated with familiar options change over time. In both cases, uncertainty increases the value of exploration. While motivational circuits facilitate exploitation of learned appetitive and aversive associations, much less is known about how these circuits contribute to explore-exploit decisions. In the present study we combined single-unit recordings of neural activity from the amygdala, ventral striatum (VS), and orbitofrontal cortex (OFC) in three rhesus macaques with computational modeling of the monkeys' behavior on a multi-arm bandit task. During the task, the monkeys learned to choose between three, probabilistically rewarded images. Periodically one of the choices was replaced with a novel image the monkey had not yet associated with reward. This explicitly induced an explore-exploit tradeoff, forcing the monkeys to choose between exploring the novel option or exploiting their existing knowledge about the two familiar options that remained available. We first used a Partially Observable Markov Decision Process model to quantify the value of choosing each option in terms of its immediate expected value (IEV), and an associated exploration bonus (BON). The IEV reflected the likelihood that a choice would be rewarded on the current trial, while the BON quantified the relative difference in the total number of future rewards to be gained by choosing to explore an option rather than exploiting alternative options (i.e. the BON is inversely related to uncertainty). We then used chosen value estimates derived from the POMDP to predict neural activity during the task. The overall representation of the IEV of the chosen option did not differ between the three brain regions, however, the representation of the chosen IEV occurred earliest in the amygdala and prior to when the animals made their choice. This contrasted with the onset

of IEV representations in VS and OFC that arose after the animals had made their choice. There was an overall stronger representation of the BON in the amygdala than in the VS and just as we saw for encoding of the chosen IEV, the onset of the chosen BON representation occurred earlier in the amygdala compared to the VS and OFC. These results imply that the amygdala plays a critical role in regulating explore-exploit decisions during reinforcement learning.

Disclosures: V.D. Costa: None. **B.B. Averbeck:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.08/TT19

Topic: H.01. Animal Cognition and Behavior

Support: NIH grant NS076416

UW RRF grant

NIA 5T32AG000057-39 grant

Title: Ventral tegmental area hypoactivity may contribute to altered age-related risk-based decision making

Authors: *V. L. TRYON, S. J. Y. MIZUMORI
Psychology, Univ. of Washington, Seattle, WA

Abstract: Normal cognitive aging can result in altered decision making behavior even in the absence of neurodegenerative disease (e.g. Fletcher & Rapp, 2013). We previously showed that aged rats exhibit increased probability discounting; that is, they decrease their choice of a risky reward when the reward is not guaranteed. This observed increase in discounting with age was partially due to a decrease in sensitivity to positive reinforcement, as measured by win-stay behavior. The goal of the current study was to assess whether age changes in midbrain ventral tegmental area (VTA) neural responses to rewards could contribute to the observed changes in probability discounting. Midbrain dopamine (DA) neurons in particular are known to be important for encoding reinforcing stimuli. DA function declines across the life-span, and this decline has been correlated with age-related cognitive deficits (Backman et al., 2006). We used a task designed to test aged (27-30 months) and young (9-12 months) rats' preference for a lever that lead to a large reward with varying probability (the 'risky' option) or a lever that lead to a small reward 100% of the time (the 'certain' option). Pressing the certain lever led to the delivery of 1 sugar pellet 100% of the time while pressing the 'risky' lever led to 4 sugar pellets with varying probability across trial blocks. The descending probabilities associated with the risky lever were 80% and 20%. Each session consisted of 4 blocks of trials, a forced choice

block followed by a free choice block for each probability condition. Each forced choice block contained 10 trials in which one lever came out at a time to inform the rat of the current reward probability. Next were 20 free choice trials in which both levers were made available, and rats were free to choose amongst the 2 options. Once rats exhibited stable performance on the task, they were implanted unilaterally with a 6 tetrode array aimed at the VTA. Analysis of the neural data revealed that putative DA neurons recorded from aged rats had significantly reduced baseline firing rate and did not respond to reward-predictive or rewarding stimuli. Additionally, aged non-DA neurons responded to reward-predictive stimuli, but there were proportionally fewer neurons recorded from aged rats relative to young rats, and the aged phasic response was significantly reduced. While the in-vivo electrophysiological data from young and aged rats' VTA are preliminary, the striking lack of dynamic range in VTA neurons of aged rats supports the view that a functional decline in these neurons may contribute to their altered risk-based decision making.

Disclosures: V.L. Tryon: None. **S.J.Y. Mizumori:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.09/TT20

Topic: H.01. Animal Cognition and Behavior

Title: Social influences on control signals in human and monkey anterior cingulate cortex (ACC) in a simulated stock market

Authors: *A. W. HUTTUNEN[1], M. L. PLATT[2]
[1]Wharton Sch., [2]CCN, Univ. of Pennsylvania, Philadelphia, PA

Abstract: Suboptimal decision-making can have devastating effects on an individual's social and financial well-being. Understanding the neural mechanisms underlying poor financial decisions - especially in social contexts - would provide critical insight into both clinical (e.g. gambling addiction, Parkinson's Disease) and healthy populations (e.g. investor 'herding' and stock market bubbles) that are adversely affected by such biases. Evidence suggests that biases such as loss aversion arise from competing valuation systems in the brain, whereby the 'low-cost' heuristic process wins out over the more effortful goal-directed computational strategy. Dorsal anterior cingulate cortex (dACC) is hypothesized to exert control over these two neurally dissociable systems by estimating the expected value of control (EVC), or the potential benefit of employing the more effortful or cognitively demanding strategy (Hayden, Pearson, & Platt, 2011; Shenhav, Botvinick, & Cohen, 2013). Given that anterior cingulate gyrus (ACCg) is commonly implicated in social value conflict monitoring and observational learning, we hypothesized a similar role for ACCg in exerting control over valuations of social information.

Based on our previous research demonstrating that rodents exhibit behavioral biases similar to humans when 'trading' with other rodents (Huttunen & Bowman, In Prep.), we developed a similar simulated stock market task to investigate whether the neural systems underpinning suboptimal social information evaluations in herding and bubbles are also evolutionarily conserved across human and nonhuman primates. Changes in the quantity and quality of the social information individuals used during the formation of market bubbles were evaluated with behavioral and eye-tracking measures in humans and monkeys. Furthermore, high resolution intracranial recordings in monkeys provides direct comparison of activity in ACCg and dACC during trading. Our model suggests that ACC plays a critical role in evaluating the amount of weight placed on social information during simulated trading decisions. Moreover, mistakes in maximizing expected value of social information, such is the case in 'herding', parallel mistakes in maximizing expected value of reward. In both cases, greater cognitive control leads to a higher proportion of optimal outcomes.

Disclosures: A.W. Huttunen: None. **M.L. Platt:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.10/TT21

Topic: H.01. Animal Cognition and Behavior

Support: Mong Neurotechnology Fellowship

NARSAD Young Investigator Award

Taiwan Ministry of Education Fellowship

Robertson Neuroscience Investigator, New York Stem Cell Foundation

NIH Director's New Innovator Award

Alfred P. Sloan Fellowship

Whitehall Foundation

Title: Environmental valence modulates dorsal raphe serotonin and GABA neural dynamics

Authors: *C. SEO[1,2], M. JIN[1], A. K. RECKNAGEL[1], E. WANG[1], C. M. BOADA[1], N. A. KRUPA[1], Y.-Y. HO[1], D. A. BULKIN[1,2], M. R. WARDEN[1,2]
[1]Neurobio. and Behavior, [2]Cornell Neurotech, Cornell Univ., Ithaca, NY

Abstract: Elevated forebrain serotonin (5-HT) has been associated with an array of inactive behavioral phenotypes that includes behavioral inhibition, learned helplessness, and the enhanced ability to wait patiently for rewards. Likewise, reduced 5-HT has been associated with active behavioral phenotypes including faster acquisition of conditioned avoidance behavior and the inability to inhibit premature responses in reward tasks. Although 5-HT neural activity during active/inactive behaviors is broadly consistent, modulation of 5-HT activity by the quality of the current environment is not well understood. Here, we investigated whether active/inactive neural dynamics in the dorsal raphe nucleus (DRN), the primary source of 5-HT to the forebrain, are modulated by environmental valence. Using fiber photometry, we recorded population activity from DRN 5-HT and GABA neurons while mice were actively behaving in rewarding and aversive environments. We first recorded neural activity while mice were subjected to the tail suspension test (poor environment), explored an open field (neutral), and were allowed to engage in free running behavior on a running wheel (good). Surprisingly, neural dynamics in dorsal raphe 5-HT and GABA populations were strongly modulated by environmental valence. 5-HT and GABA neural activity increased during struggling in the tail suspension test, did not systematically fluctuate during the open field test, and decreased during wheel running. These data suggest that environmental valence may play a role in modulating DRN neural dynamics, but leave open the possibility that gross behavioral differences might underlie this effect. We therefore subsequently tested these mice in cued approach and avoidance behaviors, in which they were required to cross a chamber either to obtain a reward or avoid a shock. When mice engaged in these visually indistinguishable running behaviors, DRN GABA neurons continued to be strongly modulated by environmental valence. DRN GABA neural activity increased during running to avoid the shock (bad environment) and decreased during running to obtain the reward (good environment), consistent with the neural dynamics observed during the tail suspension test and running wheel. DRN 5-HT neural activity was systematically suppressed during running in both good and bad environments. We are currently investigating the causal role of these neural populations in approach and avoidance behaviors with optogenetic control. These data support a major role for environmental valence in modulating dorsal raphe neural dynamics during active and inactive behaviors.

Disclosures: C. Seo: None. **M. Jin:** None. **A.K. Recknagel:** None. **E. Wang:** None. **C.M. Boada:** None. **N.A. Krupa:** None. **Y. Ho:** None. **D.A. Bulkin:** None. **M.R. Warden:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.11/TT22

Topic: H.01. Animal Cognition and Behavior

Support: Human Frontier Science Program (project RG0015/2013)

European Research Council Advanced grant, CONCEPT (project 294498)

Title: Multisensory decision making in rats and underlying neuronal mechanisms

Authors: *N. NIKBAKHT, M. ADIBI, D. ZOCCOLAN, M. E. DIAMOND
Neurosci., SISSA, Trieste, Italy

Abstract: How the brain merges information across sensory modalities to form unified percepts has been the object of debate. Here, we investigated multisensory integration in rats through an orientation categorization task. Rats encountered a grating disc composed of alternating ridges in white and grooves in black. Rats used either their vision (V), or whiskers and snout (Touch; T), or both modalities together (VT). At every trial, the grating disc was rotated to a different orientation spanning a range of 180 degrees. Rats learned to lick one spout for orientations 0±45°(horizontal) and the opposite spout for orientations 90±45° (vertical). Psychometric curves of correct responses versus the orientation showed on averaged across rats, performance was similar across V and T conditions, while individual rats had different comparative V and T performances. Irrespective of their relative V and T performances, all rats performed best in the VT condition, indicating multimodal enhancement. To quantify this enhancement, we computed the performance expected by optimal linear integration of V and T signals in the frameworks of Bayesian decision theory and information theory; both analyses revealed rats combined vision and touch better than could be accounted for by a linear interaction. High-speed video did not reveal difference in the way rats interacted with the object across modalities that may explain the supralinearity. To further investigate how the combination of sensory information from the two modalities gives rise to a decision, we trained rats in a similar task where the categorization boundary for V (at 25°) was different from that of T (65°). Two auditory tones were associated with each modality. Rats successfully learned to categorize orientations according to distinct visual and tactile boundaries, as revealed by a shift in their psychometric curves for V and T conditions. However, in VT condition, rats did not generalize the learned rule to the combination of V and T information. Extracellular recordings in the posterior parietal cortex (PPC) revealed for the majority of neurons task-related firing rate was invariant across the 3 conditions. For 251 out of 622 neurons, the firing rate was best accounted for by behavioral choice while in 185 neurons, activity was best accounted for by stimulus orientation. For the majority of neurons, orientation tuning was congruent across the 3 conditions. Neurometric curves for V, T, and VT trials were consistent with the observed higher performance on multimodal trials. Because PPC expresses activity ranging from stimulus-related to choice-related we suggest that it is involved in supramodal percept-to-choice transformation.

Disclosures: **N. Nikbakht:** None. **M. Adibi:** None. **D. Zoccolan:** None. **M.E. Diamond:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.12/TT23

Topic: H.01. Animal Cognition and Behavior

Support: NARSAD Young Investigator Award

Icahn School of Medicine at Mount Sinai Seed Funds

NIMH Grant R01MH110822

NIMH Grant R21MH112539

Title: Subcallosal anterior cingulate cortex, ventral striatum, and amygdala encode distinct aspects of reward

Authors: *M. E. YOUNG, K. D. ESANNASON, S. TAMANG, C. P. MOSHER, P. H. RUDEBECK
Dept. of Neurosci., Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Dysfunction within the circuits linking subcallosal anterior cingulate cortex (ACC), ventral striatum (VS) and amygdala is related to changes in affect, most notably anhedonia, a loss of positive affect from rewarding events. However, a mechanistic understanding of how the subcallosal ACC interacts with VS and amygdala during the anticipation and receipt of reward in normal function is lacking. Previous work has shown that lesions of the subcallosal ACC affect autonomic arousal in anticipation of rewards, suggesting that this part of the prefrontal cortex may influence reward processing in VS and amygdala during the anticipation of reward. Here we investigated how neurons in subcallosal ACC, VS, and amygdala interact while monkeys anticipate and then receive reward. Two rhesus macaques were trained to perform Pavlovian and instrumental trace conditioning tasks while single-unit and local field activity in subcallosal ACC, VS, and basolateral amygdala were recorded. Autonomic measures of arousal, such as pupil diameter and continuous EKG, and task-related behavioral responses were also continuously collected. We found that monkeys showed elevated behavioral (anticipatory licking) and autonomic (pupil size) responses in anticipation of rewards, which were modulated by each animal's individual reward preferences. Neurons in subcallosal ACC, VS, and amygdala encoded the upcoming reward during the stimulus and trace intervals, and this activity was similarly modulated by individual reward preference. However, the timing of these responses differed between areas, with amygdala neurons signaling reward value first, followed then by subcallosal ACC and last by VS. Notably, there was a large increase in the proportion of VS neurons encoding the anticipated reward just prior to reward delivery during the trace interval. In

conclusion, we found that neuronal activity in subcallosal ACC, VS, and amygdala correlates with sustained behavioral and autonomic responses in anticipation of rewards, and that these responses occur on different time scales. We are now exploring how these areas interact as a functional circuit by looking at measures of oscillatory coherence and spike-spike correlations within and between subcallosal ACC, VS, and amygdala.

Disclosures: M.E. Young: None. **K.D. Esannason:** None. **S. Tamang:** None. **C.P. Mosher:** None. **P.H. Rudebeck:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.13/TT24

Topic: H.01. Animal Cognition and Behavior

Support: CIHR (MOP 133579)

Title: Optogenetic dissection of temporal dynamics of amygdala-striatal interplay during risk/reward decision-making

Authors: *D. A. BERCOVICI[1], D. E. MOORMAN[2], M. TSE[3], S. B. FLORESCO[3]
[1]Univ. of British Colombia, Vancouver, BC, Canada; [2]Psychological and Brain Sci., Univ. of Massachusetts Amherst, Amherst, MA; [3]Univ. British Columbia, Vancouver, BC, Canada

Abstract: Assessing costs and benefits associated with different options that vary in terms of reward magnitude and uncertainty is an adaptive behaviour which motivates us to select the optimal course of action. Previous studies using reversible inactivation have shown that the basolateral amygdala (BLA) to nucleus accumbens (NAc) pathway plays a key role in promoting choice towards larger, riskier rewards. Neural activity in the BLA and NAc show distinct, phasic changes in firing prior to action initiation and following action outcomes. Yet, how temporally-precise patterns of activity within BLA-NAc circuitry influences choice behavior is unclear. We assessed how optogenetic silencing of BLA projection terminals in the NAc altered action selection. Rats that received intra-BLA infusions of AAV encoding for the inhibitory opsin eArchT were well-trained on a probabilistic discounting task, where they chose between a smaller/certain reward and a larger reward delivered in a probabilistic manner, with the odds of obtaining the larger reward changing over blocks of trials (50-12.5%). During testing, discrete 5-7s pulses of light (532 nm) were delivered via fiber optic ferrules into the NAc to suppress activity within BLA terminals during specific task events. These included periods “prior to choice” or during different “choice outcomes” (receipt of large rewards, small rewards or reward omissions). Discrete optogenetic suppression of activity in BLA inputs to the NAc prior to choice reduced selection of the more preferred option, suggesting that at this time, activity within

this pathway biases choice towards more preferred rewards. Suppression of BLA inputs during reward omissions increased risky choice during the low-probability block, indicating that activity in this circuit after non-rewarded actions serves to modify subsequent choice behavior. In contrast, silencing during rewarded outcomes did not reliably affect choice behaviour. In vivo electrophysiological studies confirmed that, in rats infused with eArchT within the BLA, local laser light application within the NAc markedly reduced firing of NAc neurons evoked by BLA stimulation. Collectively these data clarify how patterns of activity in BLA-NAc circuitry convey different types of information that guide optimal action-selection in situations involving reward uncertainty.

Disclosures: D.A. Bercovici: None. **D.E. Moorman:** None. **M. Tse:** None. **S.B. Floresco:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.14/TT25

Topic: H.01. Animal Cognition and Behavior

Support: CIHR (MOP 133579)

Title: Dorsal and ventral regions of the medial prefrontal cortex play dissociable roles in regulating risk/reward decision making during a “Blackjack” task guided by external cues

Authors: *M. VAN HOLSTEIN[1], S. B. FLORESCO[2]
[1]Psychology, Univ. of British Columbia, Vancouver, BC, Canada; [2]Univ. British Columbia, Vancouver, BC, Canada

Abstract: We often face decisions requiring a choice between options that vary in terms of reward magnitude and uncertainty. Moreover, the relative amount of uncertainty associated with different options can vary. For example, an experienced blackjack player knows the odds of winning a hand are larger when the dealer is showing a “6” compared to an “ace”. Studies with humans often present choices between taking a gamble and choosing a safe alternative, with explicit cues informing participants about the probabilities of winning a gamble prior to the decision. Functional neuroimaging studies using these tasks have revealed a role for the nucleus accumbens (NAc) and distinct parts of the prefrontal cortex (PFC), in particular the ventromedial PFC (vmPFC) and anterior cingulate (AC) cortex in risky decision making. Preclinical studies with rodents have provided additional insight in the neural basis of risk/reward decision making. The probabilistic discounting task is one common way to assess risky decision making wherein rats must use internally-generated information to identify when odds associated with each option change. To bridge this gap between assays in rats and humans we developed an operant assay we

refer to as the “Blackjack” task. In this task, animals are presented with a choice between a small/certain (1 pellet) and a large/uncertain (4 pellet) option. The odds of obtaining the large reward vary across trials, and are signaled by an auditory cue: Prior to a choice, one of two tones signalled that the odds of obtaining the larger reward were either good (50%) or poor (12.5%). Under control conditions, well-trained rats selected the large reward option more often when the odds were good vs poor (~70% vs 20%). We recently revealed distinct roles for subregions of the NAc in this form of decision making. Inactivation of the NAc shell increased risky choice when the odds were poor, suggesting it regulates suppression of suboptimal choices. Inactivation of the NAc core caused a more general disruption in using discriminative cues to guide appropriate responding. In the present study we assessed the involvement of two key cortical inputs to the NAc: the infralimbic (IL) and AC cortex. Inactivation of the IL increased risky choice in a manner similar to inactivation of the NAc shell. However, unlike inactivation of the NAc core, inactivation of the AC resulted in a decrease in the large reward option when the odds were good, suggesting it promotes selection of beneficial choices. Collectively, these data show that the IL and AC play unique roles when using cues to guide decisions associated with uncertainty, similar to the roles the vmPFC and AC might play in humans.

Disclosures: M. Van Holstein: None. **S.B. Floresco:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.15/TT26

Topic: H.01. Animal Cognition and Behavior

Support: CIHR (MOP 133579)

Title: Modulation of probabilistic discounting and reversal learning by dopamine within the medial orbitofrontal cortex

Authors: *N. L. JENNI[1], Y. T. LI[2], S. B. FLORESCO[3]
[1]Psychology, [2]Univ. of British Columbia, Vancouver, BC, Canada; [3]Univ. British Columbia, Vancouver, BC, Canada

Abstract: Weighing the value of a reward against its likelihood of delivery in order to optimize long-term utility is a necessary component of adaptive decision making. The medial subregion of the orbitofrontal cortex (mOFC) plays a key role in this form of cognition, as inactivation of this subregion in rats alters both risk/reward decision making and cognitive flexibility involving reward uncertainty (probabilistic reversal learning). The mOFC receives dopaminergic input from midbrain neurons, yet whether dopamine (DA) modulates mOFC function has been virtually unexplored. Here, we assessed how DA D_1 and D_2 receptors in the mOFC may

modulate adaptive decision making in the face of probabilistic outcomes. One series of experiments assessed risk/reward decision-making using a probabilistic discounting task, while another set of studies assessed probabilistic reversal learning. Separate groups of well-trained rats, received intra-mOFC microinfusions of selective D_1 or D_2 antagonists or agonists prior to task performance. Our results indicate that blocking mOFC D_1 receptors reduced risky choice, driven by an increase in lose-shift behavior. Interestingly, excess stimulation at the D_1 receptor did not alter choice behavior. Blockade of D_2 receptors increased, while stimulation of D_2 receptors reduced risky choice, with this reduction once again driven by increased lose-shift behavior, suggesting that mOFC DA receptors play an important role in mitigating sensitivity to non-rewarded actions to maintain optimal choice biases during risk/reward decision making. Blockade and stimulation of D_1 receptors in the mOFC reduced the number of reversals completed, but this may be accounted for by a more basic impairment in probabilistic reinforcement learning, as blockade of D_1 receptors increased errors specifically during the initial discrimination of the task. In contrast, blockade of D_2 receptors increased while stimulation of D_2 receptors reduced the number of reversals completed. Again, these effects were apparent during the initial discrimination of the task. Across both behaviours, we found that D_1 and D_2 receptors within the mOFC play dissociable and opposing roles in different forms of reward-related action selection. Together, these findings highlight a novel role for mOFC DA in guiding behavior in situations of reward uncertainty. Elucidating how DA within different nodes of mesocorticolimbic circuitry influences action selection in these situations will expand our understanding of the mechanisms regulating optimal and aberrant decision-making.

Disclosures: **N.L. Jenni:** None. **Y.T. Li:** None. **S.B. Floresco:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.16/TT27

Topic: H.01. Animal Cognition and Behavior

Support: CIHR

NSERC

Title: Local androgen synthesis in the male rat brain and its modulation of behavioral flexibility

Authors: *R. J. TOMM[1], H. R. SCHWEITZER[1], D. J. TOBIANSKY[3], G. K. KACHKOVSKI[1], C. MA[2], H. H. ADOMAT[4], E. S. GUNS[4], S. B. FLORESCO[5], K. K. SOMA[6]
[1]Psychology, [2]Univ. of British Columbia, Vancouver, BC, Canada; [3]Dept. of Psychology, The Univ. of British Columbia, Vancouver, BC, Canada; [4]The Prostate Ctr. at Vancouver Gen. Hosp., Vancouver, BC, Canada; [6]Psychology, [5]Univ. British Columbia, Vancouver, BC, Canada

Abstract: Androgens are known to regulate sexual and aggressive behavior in males. However, little attention has focused on the effects of androgens on higher-order cognition and executive function. Androgens are produced in the gonads but are also produced locally in the brain, which might be particularly important when systemic androgen levels are low. Here, we examined the effects of gonadectomy (GDX) and/or an androgen synthesis inhibitor (abiraterone acetate, ABI) on different forms of behavioral flexibility in adult male Long-Evans rats. Rats received either GDX or sham surgeries and then were housed for 5 weeks, to allow for possible upregulation of local androgen synthesis after GDX. Five days prior to the commencement of behavioral training, rats received daily treatments of either vehicle or ABI (40 mg/kg, p.o.), a compound that is used to treat patients with prostate cancer and crosses the blood-brain barrier. Behavioral flexibility was assessed using operant-based assays. Separate groups were tested on either a strategy set-shifting task or a spatial reversal learning task. The strategy set-shifting task required rats to disengage from a previously correct (but now incorrect) visual-cue based discrimination strategy, and acquire and maintain a new egocentric spatial response strategy. GDX or ABI did not affect the ability to learn an initial, visual-cue based strategy. However, during the shift to an egocentric response strategy, ABI treatment (but not GDX alone) caused an apparent improvement in behavioral flexibility, by reducing the number of errors made before reaching criterion. In a separate group of rats trained on a reversal learning task, we found a similar effect, in that only ABI reduced perseverative-type errors during the reversal. In both studies, GDX+vehicle subjects performed similarly to SHAM+vehicle subjects, suggesting that neural androgen synthesis were mediating these effects. Taken together, these data suggest that neural T synthesis may serve to increase persistence of behavior, which can in some instances supress behavioral flexibility. These data provide evidence for the behavioral functions of neural androgen synthesis in mammals. Ongoing work will measure a panel of sex steroids using LC-MS/MS in multiple brain regions that regulate behavioral flexibility. Furthermore, we will examine the effects of GDX and ABI on dopamine signaling, which plays a critical role in behavioral flexibility.

Disclosures: R.J. Tomm: None. **H.R. Schweitzer:** None. **D.J. Tobiansky:** None. **G.K. Kachkovski:** None. **C. Ma:** None. **H.H. Adomat:** None. **E.S. Guns:** None. **S.B. Floresco:** None. **K.K. Soma:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.17/TT28

Topic: H.01. Animal Cognition and Behavior

Support: NSERC

Title: Active versus passive avoidance is differentially regulated by nucleus accumbens core and shell

Authors: *P. T. PIANTADOSI[1], D. C. M. YEATES[2], S. B. FLORESCO[3]
[1]Psychology, [2]Univ. of British Columbia, Vancouver, BC, Canada; [3]Univ. British Columbia, Vancouver, BC, Canada

Abstract: Defensive behavior is an adaptive construct that enables organisms to escape or avoid potential threat. These behaviors include defensive reactions, during which behavior is suppressed in order to passively avoid a threatening situation, and defensive actions, during which behavior is produced to actively avoid danger. However, in naturalistic settings, inhibiting motor output in a context where actively avoiding may be more advantageous, or vice versa, could result in aversive consequences. Such inappropriate allocation of defensive behavior has been suggested to underlie pathological avoidance in neuropsychiatric conditions where active avoidance is abnormally elevated (e.g. depression), or response-inhibition (passive avoidance) is diminished (e.g. substance abuse). This dichotomy of active versus passive behavior may be differentially regulated by the major subdivsions of the nucleus accumbens, the core (NAcC) and shell (NAcS), which are known to control flexible action selection in the appetitive domain. Here we examined the contribution of these NAc subregions to cued active/passive avoidance behavior using a novel, operant-based Go/No-Go task. Rats were extensively trained to produce an active avoidance response (press a lever to avoid up to 3 foot-shocks over 10 s; AA) during presentation of one 15 s auditory cue, or a passive avoidance (withhold pressing for 15 s to avoid foot-shock; PA) in response to another auditory cue. Inactivation of either NAcC (n = 10) or NAcS (n = 11) (via infusion of baclofen/muscimol) impaired AA, resulting in more failures and shocks received, without altering escape behavior (i.e. lever-presses after delivery of foot-shock). In contrast, PA was disrupted by inactivation of the NAcS, but not NAcC, reflecting a deficit in response-inhibition that manifested as more passive avoidance failures, lever-presses during a passive failure, and locomotion. To examine the contribution of monoaminergic signaling to AA/PA performance, high-performing rats were administered a systemic dose of D-amphetamine (1 mg/kg). This dose did not affect AA performance, but disinhibited behavior, increasing PA failure (but leaving the number of passive presses intact) and locomotion, suggesting that rats remained sensitive to the instrumental punisher despite behavioral disinhibition. These results provide insight into the ventral striatal regulation of complex avoidance behavior, supporting previous work suggesting that the NAcS is part of an AA circuit, and adding to a growing literature indicating that the NAcS inhibits inappropriate or punished behavior, while the NAcC promotes flexible approach behavior.

Disclosures: P.T. Piantadosi: None. **D.C.M. Yeates:** None. **S.B. Floresco:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.18/TT29

Topic: H.01. Animal Cognition and Behavior

Support: FEDER funds, through the Competitiveness Factors Operational Programme (COMPETE)

Foundation for Science and Technology (FCT) Project: POCI-01-0145-FEDER-007038

FCT Project: PTDC/NEU-SCC/5301/2014

FCT Grant: SFRH/BD/52291/2013 (ME via Inter-University Doctoral Programme in Ageing and Chronic Disease, PhDOC)

FCT Grant: SFRH/BD/109111/2015 (AMC)

FCT Grant: PDE/BDE/113602/2015 (JSR)

FCT Grant: SFRH/BPD/80118/2011 (HA)

Title: Functional encoding of inhibitory control in rats in the variable delay-to-signal task

Authors: *M. ESTEVES, A. M. CUNHA, J. S. REIS, A. ALMEIDA, N. SOUSA, H. LEITE-ALMEIDA
ICVS, Sch. of Medicine, Univ. of Minho, Braga, Portugal

Abstract: The nucleus accumbens (NAcc) is widely known to be associated with critical aspects of decision-making, namely motivation and reward. Although not completely clear, this region has also been linked to behavioral inhibition, for which associations with the prefrontal cortex seem to have an important role. NAcc oscillatory activity was registered in Wistar-Han rats implanted with nichrome single wire electrodes during the execution of an impulsivity task - Variable Delay-to-Signal (VDS). During 10 training sessions animals learnt to nosepoke a small aperture after a 3s delay (signaled with a light) to earn a sugared pellet. Premature nosepokes (during the delay) were punished with a timeout and no reward was provided. On the last session (test), premature responses were allowed (i.e. not punished) and delays were variable. The test started with 25 trials of 3s delays, followed by 70 trials of variable (6 and 12s) delays and finally the 25 trials with 3s delays were repeated. While the training construct reflects action impulsivity, the VDS test is more akin to delay tolerance and correlates with the delay discounting behavior. Local Field Potentials (LFPs) were acquired during the task performance,

amplified and band-pass filtered. Processing was performed in Matlab using the Chronux toolbox and included downsampling, referencing, artifact removal, event alignment, power estimation using a multitaper method and averaging. NAcc power showed frequency-dependent variations around the time of nosepoke, particularly an increase in the lowest frequencies. This activity peak preceded the premature responses but was simultaneous with the timed responses. Furthermore, power in high frequencies was elevated approximately 750ms before premature responses. Our results indicate that the activity state in the Nacc conditions animals' inhibitory capacity suggesting that this area is critically involved in impulsive behavior.

Disclosures: M. Esteves: None. **A.M. Cunha:** None. **J.S. Reis:** None. **A. Almeida:** None. **N. Sousa:** None. **H. Leite-Almeida:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.19/TT30

Topic: H.01. Animal Cognition and Behavior

Support: POCI-01-0145-FEDER-007038

PTDC/NEU-SCC/5301/2014

SFRH/BD/109111/2015

SFRH/BD/52291/2013

PD/BD/114117/2015

SFRH/BPD/80118/201

Title: Opposite role of left and right nucleus accumbens in impulsive and habit to goal-directed decision-making

Authors: *A. FERREIRA CUNHA, M. GUIMARÃES, M. ESTEVES, F. TEIXEIRA, A. SALGADO, A. RODRIGUES, N. SOUSA, A. ALMEIDA, H. LEITE-ALMEIDA
Life and Hlth. Sci. Res. Inst. (ICVS), Braga, Portugal

Abstract: Nucleus accumbens (NAcc) is highly involved in impulsive and habit-based/goal-directed decision-making. Curiously, such involvement appears to be lateralized, particularly in the case of impulsivity. Indeed, decreased expression of glutamate decarboxylase, markers of dendritic spines and microtubules, as well as decreased gray matter density in the left (but not in the right) NAcc were associated with high trait impulsivity (Caprioli et al 2014). In this study, we analyzed if the depletion of dopaminergic efferences to the left and right NAcc affected

distinct aspects of decision-making. Wistar-Han male rats, 8 weeks-old, received an injection of 6-hidroxydopamine in the left (6-OHDA-L) or right (6-OHDA-R) NAcc (AP: 1.2, ML: +/- 2.1, DV: 7.0). Control animals were injected with vehicle in the same coordinates. After 3 weeks, rats were trained in an operant chamber to perform an action (lever press) contingent with an outcome (reward). The action/outcome ratio was sequentially increased across sessions up to a random ratio of 20 lever presses/outcome (RR20). An early devaluation session was performed at this stage. In the following 6 sessions, animals were trained in a RR20 schedule (habit formation) and finally a late devaluation test was performed. Two more RR20 sessions were done, in the end of which a contingency of one of the levers was degraded for 2 sessions. Animals performed 3 extra sessions of RR20 in the non-degraded lever and in the end a devaluation test. Then animals were tested in the Variable-delay-to-signal test (VDS; Leite-Almeida et al. 2013) for impulsive behaviour. In a second experiment, a group of animals performed only the VDS test. No differences were found between groups in the learning phase of both operant tasks. However, 6-OHDA R (contrary to Sham or 6-OHDA L) animals do not degraded the contingency in the first day of the test neither devaluate the reward when only one lever was present, indicating a impairment in the shift from habit towards goal-direct behaviour in this group. In opposition, in the VDS test, 6-OHDA-L group manifested higher delay intolerance, as observed by a rampant accumulation of premature responses during the large delay trials (6 and 12 s), than 6-OHDA-R or Sham animals. Additional tests demonstrate that these alterations are not the result of impaired motivation, anxiety-like behavior nor motor problems. Our results demonstrate an opposite involvement of the NAcc in the manifestation of impulsive and goal-directed behaviors and that such is associated with a differential contribution of the left and right mesolimbic dopaminergic projections. Follow up studies will assess the specific involvement of D1 or D2 cells.

Disclosures: **A. Ferreira Cunha:** None. **M. Guimarães:** None. **M. Esteves:** None. **F. Teixeira:** None. **A. Salgado:** None. **A. Rodrigues:** None. **N. Sousa:** None. **A. Almeida:** None. **H. Leite-Almeida:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.20/TT31

Topic: H.01. Animal Cognition and Behavior

Support: OFT

NIMH

NIH

Title: Visual two-arm bandit reinforcement learning in rodents

Authors: *G. M. ZABALA-ALEMAN[1,2,3], J. DU HOFFMANN[1,4,5], Y. CHUDASAMA[1,4,5], B. B. AVERBECK[1,2]
[1]Natl. Inst. of Mental Hlth., Bethesda, MD; [2]Unit on Learning and Decision Making, Bethesda, MD; [3]Univ. Central del Caribe, Bayamon, Puerto Rico; [4]Section on Behavioral Neurosci., Bethesda, MD; [5]Rodent Behavioral Core, Bethesda, MD

Abstract: Reinforcement learning (RL) is the behavioral process of learning to make choices to gather rewards and avoid losses. RL is often studied in the context of two-armed bandit tasks, in which subjects have to make choices between a pair of options. After each choice, they receive an outcome. By integrating the outcomes over trials they can learn which choice is most rewarding, and choose that option in the future. In rodents, choices in such tasks are often between levers or ports in different locations within an operant box. The neural circuitry that underlies learning about which port or which lever is most rewarding has been examined. In studies using primates and humans, choices are often between visual objects whose location varies trial-by-trial. Recent evidence suggests that the neural circuitry that underlies learning to select rewarding actions is different than the neural circuitry that underlies learning to select rewarding objects. Therefore we developed a rodent two-armed bandit task using a touchscreen platform in which rodents (N = 12 rats) initiated a trial by nose-poking a food magazine located opposite the visual display. Two different geometric objects were presented on the left and right side of the touchscreen. The animals then approached the touchscreen and made a choice by nose-poking one of the two visual objects. A response to the correct object was rewarded with 10% sucrose liquid reward. A response to the incorrect object was not rewarded. On average, the animals acquired the stimulus-reward association within 5 days, selecting the rewarded object more than 80% of the time. We subsequently reversed the stimulus-reward contingencies such that the animal had to reverse their preference. The animals successfully reversed their preference within an average of 9 days and switched to selecting the newly rewarded object. Following the initial reversal, the animals were then subjected to three additional reversals using the same pair of visual objects. The animals took an average of 7 days to reverse. Future experiments will examine the neural circuitry underlying these behaviors.

Disclosures: G.M. Zabala-Aleman: None. **J. du Hoffmann:** None. **Y. Chudasama:** None. **B.B. Averbeck:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.21/TT32

Topic: H.01. Animal Cognition and Behavior

Support: Wellcome Trust Strategic Award (100202/Z/12/Z), DEFINE – Defining Endophenotypes From Integrated Neuroscience

Medical Research Council Centre for Neuropsychiatric Genetics and Genomics (MR/L010305/1)

School of Psychology, Cardiff University

School of Medicine, Cardiff University

Title: Behavior-dependent limbic-cortical interactions in a Cyfip1 knockout rat model of psychiatric risk

Authors: *J. HECKENAST[1,3], S. TRENT[2], J. HALL[2], L. WILKINSON[2], M. W. JONES[3]
[2]Neurosciences & Mental Hlth. Res. Inst., [1]Cardiff Univ., Cardiff, United Kingdom;
[3]Physiology, Pharmacol. and Neurosci., Univ. of Bristol, Bristol, United Kingdom

Abstract: Cyfip1 gene dosage is reduced in 15q11.2 deletion syndromes, which are associated with a range of developmental, intellectual and psychiatric symptoms. While the neural bases of these symptoms are unknown, abnormal limbic-cortical interactions feature in other psychiatric patients and genetic risk factor carriers, in other rodent models, and are increasingly thought to contribute to cognitive deficits. We are using a novel line of Cyfip1 heterozygous knockout rats (Cyfip1±) to define the consequences of Cyfip1 knockdown for brain function and behavior.Eight wildtype (WT) and 5 Cyfip1± littermates were trained on a rewarded alternation T-maze task invoking working memory, a process consistently impaired in psychiatric disorders. Chronically implanted microelectrode arrays were used to simultaneously record local field potentials (LFP) from dorsal and ventral CA1 of the hippocampus (CA1), medial prefrontal cortex (PFC), nucleus accumbens and amygdala during maze performance and subsequent sleep.Choice accuracy on the task was unimpaired in Cyfip1± rats (88±5%, WT 79±4%; $p>0.05$), and LFP spectral profiles also appeared normal, showing CA1 and PFC 5-10Hz theta power similar to WT. This suggests that local networks remain broadly intact in Cyfip1± rats.However, using spectral coherence to infer functional interactions between CA1 and PFC indicated that theta coherence in Cyfip1± rats was more sensitive to cognitive context than in WT. The difference in coherence between sample and choice phases on the T-maze was significantly higher in Cyfip1± than WT ($p<0.05$). This result is reminiscent of an fMRI study in which schizophrenia risk allele carriers showed increased limbic-cortical coupling, with no impact on task performance[1].Introducing a 5min delay to the task reduced choice accuracy to chance levels and abolished the context dependence of CA1-PFC coherence in both groups, suggesting that rats may no longer engage CA1-PFC under these conditions. Current analyses are quantifying CA1-PFC interactions during sleep in Cyfip1± rats.These findings indicate that impaired neural network activity during a working memory task is a consequence of reduced Cyfip1 gene dosage, and may be an important component of the pathophysiology underlying psychiatric conditions.1. Esslinger et al, Science 2009

Disclosures: **J. Heckenast:** None. **S. Trent:** None. **J. Hall:** None. **L. Wilkinson:** None. **M.W. Jones:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.22/TT33

Topic: G.03. Emotion

Support: Swedisch Research Council

Brazilian Research Council

Olle Enqvist post doc fellowship

Title: Ventral hippocampus olm cells control type 2 theta oscillations and response to predator odor

Authors: *S. MIKULOVIC[1], E. RESTREPO[2], S. SIWANI[2], A. B. TORT[3], K. KULLANDER[2], R. N. LEAO[4]
[1]Uppsala University, Neurosci. Dept., Uppsala, Sweden; [2]Uppsala Univ., Uppsala, Sweden; [3]Brain Institute, UFRN, Natal, Brazil; [4]Brain Inst., UFRN, Natal, Brazil

Abstract: The dorsal and ventral hippocampus exert cognition and emotion-related functions, respectively. Since both regions display rhythmical activity, specific neural oscillatory pacemakers could underlie their functional dichotomy. Type 1 theta oscillations are strongly observed in the dorsal hippocampus during movement and exploration and are independent of cholinergic transmission. In contrast, type 2 theta depends on acetylcholine and appears when animals are exposed to emotionally laden contexts (e.g. predator odor tests). However, despite its involvement in emotions, the source of type 2 theta has never been attributed to the ventral hippocampus. Here we show that optogenetic activation of oriens-lacunosum moleculare (OLM) interneurons in the ventral hippocampus drives type 2 theta oscillation. We also show that OLM-driven type 2 theta oscillations can coexist with type 1 theta oscillation. Moreover, we found that type 2 theta generation is associated with increased risk-taking behavior in response to predator odor. These results demonstrate that two theta oscillation subtypes predominate in different hippocampal regions that underlie distinct cognitive functions.

Disclosures: S. Mikulovic: None. **E. Restrepo:** None. **S. Siwani:** None. **A.B. Tort:** None. **K. Kullander:** None. **R.N. Leao:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.23/TT34

Topic: G.03. Emotion

Support: KAKENHI 16H04652

Title: The macaque medial prefrontal cortex neurons respond rapidly and strongly to images of snakes and emotional faces of conspecifics

Authors: *H. NISHIMARU, H. TRONG DINH, J. MATSUMOTO, Y. TAKAMURA, Q. VAN LE, E. HORI, T. ONO, H. NISHIJO
Syst. Emotional Sci., Grad. Sch. Med. Pharm. Sci., Univ. Toyama, Toyama-shi, Japan

Abstract: Primates devote considerable attention to snakes and conspecific faces. However, neuronal mechanisms of rapid detection of such biological stimuli are still unknown in primates. It has been shown that subcortical visual system including the superior colliculus, pulvinar and amygdala, provide the neural substrate that allows rapid visual detection of snakes and emotional faces. In our previous study, we have shown that monkey pulvinar neurons respond more quickly and more strongly to snakes and faces than to other stimuli (Le et al. PNAS 2013, Nguyen et al. Eur J Neurosci 2013). The medial pulvinar and amygdala send robust inputs to the medial prefrontal cortex (mPFC), a cortical area that has been implicated in the allocation of attention to biologically important stimuli. These neural connections suggest that the mPFC is also involved in the rapid detection of such stimuli. In the present study, we report that monkey mPFC neurons responded more rapidly and more strongly to snakes than to other potential predators, and to emotional faces of conspecifics than to neutral faces of conspecifics and faces of humans. Neuronal responses in the monkey mPFC were recorded while monkeys discriminated 8 categories of visual stimuli (snakes, monkey faces, human faces, raptors, carnivores, non-predators, monkey hands, simple geometrical patterns). Of 538 mPFC neurons recorded, 93 responded to the visual stimuli. The responses to snakes and monkey faces were unique in that; 1) the ratios of neurons that responded best to snakes and monkey faces were larger than other categories, 2) mean response latencies were faster to snakes and monkey faces than those to other categories, and 3) neuronal responses to snakes were unaffected by low pass filtering of the images, but were decreased by high pass filtering. These results provide neurophysiological evidence that the mPFC is involved in coarse and rapid visual information processing of snakes and monkey faces. Furthermore, they suggest that ecological and social selective pressures shaped the PFC, a cortical area that has greatly expanded in primates.

Disclosures: **H. Nishimaru:** None. **H. Trong Dinh:** None. **J. Matsumoto:** None. **Y. Takamura:** None. **Q. Van Le:** None. **E. Hori:** None. **T. Ono:** None. **H. Nishijo:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.24/TT35

Topic: G.03. Emotion

Support: Medical Research Grant

Title: Tactile stimulation of the face and body elicit neural activity in the monkey amygdala

Authors: *J. MORROW[1], C. P. MOSHER[2], K. M. GOTHARD[3]
[1]Neurosci., Univ. of Arizona, Tucson, AZ; [2]Advanced Hlth. Sci., Cedars Sinai Med. Ctr., Los Angeles, CA; [3]Univ. Arizona, Col. Med., Tucson, AZ

Abstract: Recent studies from our laboratory reported the presence of neurons in the monkey amygdala that respond to tactile stimuli. The majority of tactile neurons had bilateral, composite receptive fields (i.e., a receptive field covering multiple, non-contiguous areas on both sides of the face). In this initial report, only the face was tested for tactile responsiveness. The restriction of tactile stimulation to only the face leaves questions about how (or if) the primate amygdala represents the rest of the body. To address this issue, we designed a solenoid controlled air puff apparatus that delivered highly controlled innocuous tactile stimulation via small nozzles to 16 different areas of the face and body. The nozzles were aimed bilaterally to the lower muzzle, upper muzzle, brow, ear, neck, upper forearm, lower forearm, and the back of the hand. Air puffs of 1 s duration were delivered to each location 15-20 times at a pressure of approximately 0.14 MPa. Neurons in the amygdala responded to tactile stimulation both on the face and body. Of the 37 neurons recorded, 7 responded to stimulation of the face while 6 responded to air puffs delivered to different parts of the body. Unlike neurons typically seen in primary somatosensory cortex, some tactile neurons in the amygdala exhibited composite receptive fields spanning distant locations on the head and body (e.g., brow and hand). While a majority of tactile receptive fields on the face and neck were symmetrical (7/10) we found no bilateral receptive fields on the limbs. Ongoing experiments will determine whether neural responses to touch on the body and limbs differ from touch on the face and whether there are significant differences in the properties of the receptive fields for these regions (e.g., size, symmetry, and continuity). These data support the idea that the amygdala is involved in processing touch stimuli regardless of where on the body the stimulation occurs.Supported by a Medical Research Grant from an Anonymous Foundation

Disclosures: J. Morrow: None. **C.P. Mosher:** None. **K.M. Gothard:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.25/TT36

Topic: G.03. Emotion

Support: Medical Research Grant

Title: Neurons in the primate amygdala respond to tactile, auditory, and visual stimuli

Authors: *P. E. ZIMMERMAN[1], J. MARROW[1], C. P. MOSHER[2], K. M. GOTHARD[3]
[1]Univ. of Arizona, Tucson, AZ; [2]Cedars-Sinai Med. Ctr., Los Angeles, CA; [3]Univ. Arizona, Col. Med., Tucson, AZ

Abstract: The primate amygdala receives input from all sensory modalities; however, the majority of neurophysiological studies in primates are limited to the domain of vision. A smaller number of studies showed that auditory stimuli also elicit responses from neurons in the primate amygdala. Recently, our laboratory reported neurons in the amygdala that responded to tactile stimuli. Given that the amygdala receives inputs from areas with different levels of sensory integration, we sought to assess the extent to which neurons in the amygdala respond to tactile, visual, and auditory stimuli. We recorded the responses of single units in the amygdala to neutral stimuli across these three domains. Only neutral, non-social stimuli were used in order to eliminate possible confounds with responses elicited by the valence/value or social salience of stimuli. Of 103 recorded neurons, 28 cells responded to a single modality, while 16 and 7 cells responded to two and three sensory modalities respectively. The most common multisensory neurons responded to visual and tactile stimuli (9 cells). Fewer neurons responded to visual-auditory (3) and auditory-tactile (4) stimulus combinations. Albeit preliminary, these data indicate that a large subset of neurons in the amygdala are multisensory. Ongoing experiments will compare responses to neutral stimuli with responses elicited by stimuli of species-specific, socio-emotional value (such as facial expressions, social calls, and grooming). These data will help establish the role of the amygdala in integrating unconditioned stimuli with low or high socio-emotional value across sensory domains.
Supported by a Medical Research Grant from an Anonymous Foundation

Disclosures: P.E. Zimmerman: None. **J. Marrow:** None. **C.P. Mosher:** None. **K.M. Gothard:** None.

Poster

423. Information Processing, Decision Making, and Reinforcement

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 423.26/TT37

Topic: G.03. Emotion

Support: Silvio O. Conte Center P50MH100023

Title: Oxytocin (intranasal or bilateral microinjections) enhances the selectivity of amygdala neural responses in a social discrimination task

Authors: *P. PUTNAM[1,2], P. E. ZIMMERMAN[2], L. J. YOUNG[3], K. M. GOTHARD[2,2]
[1]Neurosci., [2]Dept. of Physiol., Univ. Of Arizona, Tucson, AZ; [3]Dept. of Psychiatry and Behavioral Sci., Emory Univ., Atlanta, GA

Abstract: The amygdala in primates is crucial for the evaluation of social stimuli, and contains neural specializations for the individuation of conspecifics, the discrimination of facial expressions, and the detection of important social signals such as eye contact. Previous studies have shown that unilateral injections of Oxytocin (OT), a hormone and neuromodulator, into the primate basolateral amygdala increased the frequency of pro-social decisions and attention to a social partner[1]. It has also been reported that intranasal administration of OT increases blood flow in the amygdala of humans performing social tasks[2]. Despite numerous reported behavioral effects of OT, little is known about the underlying neural mechanisms by which OT modulates social cognition. To investigate the effect of OT administration on neural responses in the primate amygdala we recorded neurons from a monkey who received OT (or vehicle control) through intranasal nebulization, or direct bilateral microinjections into the amygdala. During recording the monkey discriminated between the identities of conspecific monkeys, presented in videos. Videos of objects were used as non-social controls. Using the depth of selectivity index[3] we found that intranasal administration of OT increased the selectivity of amygdala neurons for monkey identity when compared to saline inhalation ($p=0.0135$ using a Wilcoxon rank sum test). Ongoing experiments will determine whether intra-amygdala microinjection of OT can yield similar results and whether the effect of OT on social behavior can be localized to specific neuroanatomical substrates. *This work was supported by the Silvio O. Conte Center For Oxytocin and Social Cognition P50MH100023* 1. Chang SW, Fagan NA, Toda K, Utevsky AV, Pearson JM, Platt ML. Neural mechanisms of social decision-making in the primate amygdala. Proc Natl Acad Sci USA. 2015;112(52):16012-7. 2. Domes G, Heinrichs M, Gläscher J, Büchel C, Braus DF, Herpertz SC. Oxytocin attenuates amygdala responses to emotional faces regardless of valence. Biol Psychiatry. 2007;62(10):1187-90. 3. Rainer G, Asaad WF, Miller EK. Selective representation of relevant information by neurons in the primate prefrontal cortex. Nature. 1998;393(6685):577-9.

Disclosures: **P. Putnam:** None. **P.E. Zimmerman:** None. **L.J. Young:** None. **K.M. Gothard:** None.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.01/TT38

Topic: H.01. Animal Cognition and Behavior

Support: NSERC

Title: Stable changes in H2A.Z binding and acetylation during memory formation and maintenance

Authors: ***K. NARKAJ**[1], A. AZAM[1], A. ANGCO[2], K. SERVADO[2], I. B. ZOVKIC[3]
[1]Cell and Systems Biol., [3]Psychology, [2]Univ. of Toronto Mississauga, Mississauga, ON, Canada

Abstract: Memory formation is a protracted process that initially involves the hippocampus and becomes increasingly dependent on the cortex as the memory ages. Existing research has implicated stable changes in DNA methylation as a contributor to this process, whereas changes in histone modifications are typically found to be transient. Here, we investigated whether histone H2A.Z, a newly identified epigenetic regulator of learning and memory, is stably modified in the hippocampus and the cortex at delayed time points after learning. Mice were exposed to contextual fear conditioning and brains were collected either 24 hours or 30 days after learning. Using chromatin immunoprecipitation, we quantified H2A.Z and acetylated H2A.Z (AcH2A.Z; a positive marker of transcription) binding on memory-related genes. After 24h, we observed increased levels of H2A.Z acetylation on immediate early genes in fear conditioned mice in the hippocampus, demonstrating the persistence of this epigenetic modification beyond the initial consolidation window (up to 6h). 30 days after training, we detected lasting changes in both H2A.Z binding and H2A.Z acetylation in the medial prefrontal cortex (mPFC), particularly on synapse-related genes. Previously, we showed that H2A.Z in both the hippocampus and the cortex is transiently modified after learning. Here, we show that H2A.Z is stably regulated, but that this regulation is gene-specific, such that changes associated with immediate-early genes are no longer evident at 30 days, whereas, changes associated with synaptic genes persist to support memory maintenance.

Disclosures: **K. Narkaj:** None. **A. Azam:** None. **A. Angco:** None. **K. Servado:** None. **I.B. Zovkic:** None.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.02/TT39

Topic: H.01. Animal Cognition and Behavior

Support: NRF-2012R1A3A1050385

POSCO TJ Park Foundation

Title: 3D chromatin organizer, CCCTC-binding factor (CTCF), regulates remote memory and cortical synaptic plasticity

Authors: *S. KIM[1], N.-K. YU[5], J.-I. KIM[2], K.-W. SHIM[1], D. CHOI[1], S.-W. LEE[3], J. CHOI[1], J.-H. LEE[6], B.-K. KAANG[4]
[1]Seoul Natl. Univ., Seoul, Korea, Republic of; [2]Seoul Natl. Univ., Seoul-City, Korea, Republic of; [3]Biol. Sci., [4]Departments of Biol. Sci., Seoul Natl. Univ., Seoul, Korea, Republic of; [5]Scripps, LA Jolla, CA; [6]Kyung Hee Univ., Seoul, Korea, Republic of

Abstract: Molecular mechanism of long-term memory has been extensively studied in the context of hippocampus-dependent recent memory examined within several days; however, months-old remote memory maintained in the cortex for long-term has not much been investigated at molecular levels yet. Various epigenetic mechanisms are known to be important for long-term memory, but whether and how 3D chromatin architecture contributes to neuronal plasticity and memory consolidation are largely unknown. To assess memory upon perturbation of crucial 3D chromatin controller, here we utilized the conditional knockout (cKO) mice, in which CCCTC-binding factor (CTCF) is lost in neurons during adulthood. This enabled us to circumvent the lethal effect of CTCF deletion during the embryonic or postnatal development. Our CTCF cKO mice were viable for more than 8 months and showed normal recent memory in contextual fear conditioning and spatial water maze task. However, they surprisingly showed remarkable impairments in remote memory in both tasks. Underlying the remote memory-specific phenotypes, we found that loss of CTCF disrupts cortical long-term potentiation (LTP) but not hippocampal LTP. CTCF knockdown in cultured cortical neurons altered the expression of hundreds of genes, some of which we uncovered to be regulated by neuronal activity. These results suggest that remote memory storage in the cortex requires CTCF-mediated chromatin regulation in neurons while recent memory formation in the hippocampus does not.

Disclosures: S. Kim: None. **N. Yu:** None. **J. Kim:** None. **K. Shim:** None. **D. Choi:** None. **S. Lee:** None. **J. Choi:** None. **J. Lee:** None. **B. Kaang:** None.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.03/TT40

Topic: H.01. Animal Cognition and Behavior

Support: R01 MH087463

Title: Role of nuclear receptor Nr4A in hippocampus dependent memory formation: A single-cell RNA seq approach

Authors: *S. CHATTERJEE[1], M. E. GAINE[1], E. BAHL[2], J. J. MICHAELSON[2], T. ABEL[1]
[1]Dept. of Mol. Physiol. and Biophysics, [2]Dept. of Psychiatry, The Univ. of Iowa, Iowa City, IA

Abstract: New experiences are initially encoded as labile short-term memories, which are converted into stable long-term memory by gene transcription-dependent processes. Gene expression after learning involves a transient wave of transcription that is critical for memory consolidation. Following early transcriptional activity, learning also induces persistent long-lasting transcriptional changes that are reported to be involved in the storage of long-term memory. Increased transcript levels of 13 nuclear receptors, including all 3 members of the Nr4A subfamily, have been identified after learning. In this study, we show that transgenic mice expressing a dominant-negative form of NR4A (Nr4ADN) in forebrain neurons are impaired in long-term spatial memory consolidation. RNA sequencing from dorsal hippocampus after spatial object recognition task shows downregulation of genes related to long-term spatial memory and glutamate receptors. Differential splice variants, miRNA and long non-coding RNA expression is presently been analyzed. We are also using single cell RNA seq approach to measure gene expression from specific CA1 neurons responsive to learning from control and Nr4ADN mice. To measure mutant Nr4A occupancy on gene promoters, we are using YFP sorted cells (Nr4ADN fused with YFP) from dorsal hippocampus to perform X-ChIP and CUT & RUN. Therefore, the present study investigates the mechanism of Nr4A function and further underscores the importance of Nr4A during hippocampus dependent memory storage.

Disclosures: S. Chatterjee: None. **E. Bahl:** None. **M.E. Gaine:** None. **J.J. Michaelson:** None. **T. Abel:** None.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.04/TT41

Topic: H.01. Animal Cognition and Behavior

Support: National Natural Science Foundation of China 81460216

National Basic Research Program of China 2013CB835103

Strategic Priority Research Program of the Chinese Academy of Science XDB02020002

Title: Hippocampal 5-HT2A receptors regulate Rac1 activity for forgetting of contextual fear memory

Authors: *L. JIANG[1], Y. YIN[2], R. MAO[3], L. XU[4]
[1]Dali university, Yunnan, China; [2]The third People's hospital of Yunnan province, Kunming, China; [3]Key Lab. of Animal Models and Human Dis. Mechanisms, [4]Kunming Inst. of Zoology, the Chinese Acad. of Sci., Kunming, China

Abstract: Excessive fear can lead to mental disorders such as phobias and posttraumatic stress disorder. Serotonin is an ancient neurotransmitter, involved in modulating the acquisition of conditioned emotional memory such as fear and anxiety. The serotonin 2a receptor (5-HT2aR) may be one of the important postsynaptic targets to mediate such effects. Here, we investigated whether activation of hippocampal 5-HT2aR in rat could contribute to the hypermnesia of contextual fear. We found that spaced but not massed training of contextual fear conditioning caused up-regulation the expression of 5-HT2aR in the hippocampus and heightened contextual fear. Intrahippocampal injection of the 5-HT2aR antagonist MDL11939 weakened contextual fear in spaced training, while 5-HT2aR agonist TCB-2 heightened contextual fear in massed training rats. Furthermore, we found that TCB-2 induced obvious inhibition of Rac1 activity in the hippocampus. Our study firstly demonstrates that contextual fear memory in rats is actively regulated by 5-HT2aR in the hippocampus. We propose that forgetting of contextual fear is modulated by counteracting components of a molecular pathway involving 5-HT2aR, Rac1 and cofilin. Our data suggest that this pathway could provide a suitable target for therapeutic treatment of emotional disorders

Disclosures: **L. Jiang:** None. **Y. Yin:** None. **R. Mao:** None. **L. Xu:** None.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.05/TT42

Topic: H.01. Animal Cognition and Behavior

Support: Roth Foundation

FiveX

ISG Foundation

Walter and Edith Sheldon

Patricia A Quick Foundation

Title: Association between completeness of context memory formation and the number of cells incorporated into hippocampal neuronal ensembles after learning

Authors: *J. A. LEAKE[1], R. ZINN[3], L. H. CORBIT[2], B. VISSEL[3]
[1]The Univ. of Sydney, Sydney University, Australia; [2]The Univ. of Sydney, Sydney, Australia; [3]The Univ. of Technol. Sydney, Sydney, Australia

Abstract: In a contextual fear conditioning paradigm, animals require a minimum period of time in the context prior to shock to display substantial conditioned fear at test. This period is thought to be necessary for animals to form an integrated representation of the context, which then comes into association with the shock. According to this model, longer placement shock intervals support the generation of a more complete contextual representation within the hippocampus. Yet, the neural underpinnings of this increasing completeness have yet to be systematically investigated. In the present study, we tested the hypothesis that the placement shock interval controls memory completeness by limiting the number of cells activated and incorporated into the memory circuit during learning. Consistent with this, we found that longer intervals were associated with increasing numbers of immediate-early gene expressing cells within the hippocampus across an extensive PSI range. This suggests that the hippocampus continues encoding information many minutes into a learning session, and that the number of cells activated during learning may be a correlate of the completeness of memory acquisition.

Disclosures: **J.A. Leake:** None. **R. Zinn:** None. **L.H. Corbit:** None. **B. Vissel:** None.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.06/TT43

Topic: G.06. Post-traumatic Stress Disorder

Support: CAPES

CNPq

Title: Up-regulation of dorsal hippocampal kappa-opioid receptors modulate fear memory intensity during consolidation in rats

Authors: *T. C. DE LIMA[1], F. VANZ, 88040-970[2], M. A. BICCA[3], M. GIACHERO[2], L. J. BERTOGLIO[2]
[1]Univ. Federal de Santa Catarina, Florianopolis, Brazil; [2]Pharmacol., Federal Univ. of Santa Catarina, Florianopolis, Brazil; [3]Neurobio., Northwestern Univ., Evanston, IL

Abstract: Kappa-opioid receptors (KORs) are expressed in brain regions involved in aversive memory consolidation, including the dorsal hippocampus (DH). The present study sought to investigate the contribution of DH KORs to contextual fear memory consolidation. Male Wistar rats were conditioned to the context A with one shock (0.7 mA) and received a bilateral infusion into the DH of vehicle (VEH) or the selective KOR antagonist *nor*-Binaltorphimine (*nor*-BNI 1, 3 or 10 nmol/hemisphere) 0 or 6 h later. In certain experiments, *nor*-BNI was infused immediately after an unpaired shock (immediate shock). Test session was performed one (test A1) and seven (test A2) days after the conditioning day. Freezing behavior was measured as an index of memory retention. In the KORs immunoblotting analysis, animals were conditioned (except the *naive* group) and the DH was dissected 0, 1, 3 or 6 h later. Finally, animals were conditioned (context-shock pairing) or just exposed to contextual (context no-shock) or aversive (immediate shock) components, and the DH was dissected 1 h later. In experiment 1, infusion of *nor*-BNI 3 or 10 nmol immediately after conditioning increased freezing time in both tests A1 and A2 when compared with respective VEH groups (Test A1: VEH = 40 + 5%, *nor*-BNI 3 = 80 + 5%, *nor*-BNI 10 = 68 + 6%; Test A2: VEH = 36 + 4%, *nor*-BNI 3 = 69 + 3%, *nor*-BNI 10 = 65 + 4%). Moreover, *nor*-BNI 3 nmol infused 6 h after the conditioning session (experiment 2) or immediately after the immediate shock (experiment 3) had no effect on freezing behavior ([VEH = 38 + 9% vs. *nor*-BNI 3 = 39 + 6%] and [VEH = 11 + 2% vs. *nor*-BNI 3 = 12 + 2%], respectively). In experiment 4, contextual conditioning induced an increase in DH KORs immunoblotting content in groups 1 and 3 h when compared with *naive* or 0 h groups (Naive = 100 + 11%, 0 h = 101 + 12%, 1 h = 163 + 21%, 3 h = 171 + 17%). Finally, in experiment 5, shock-context group, but not context or immediate shock groups, showed DH KORs immunoblotting content increased 1 h after training session (Naive = 100 + 13%, Context no-

shock = 147 + 13%, Context-shock pairing = 171 + 24%, Immediate shock = 114 + 8%). Altogether, these results suggest that DH KORs are up-regulated during an associative aversive experience, with their activation playing an important modulatory role in contextual fear memory consolidation. Financial support: CAPES, CNPq

Disclosures: **T.C. de Lima:** A. Employment/Salary (full or part-time):; Federal University of Santa Catarina - Department of Pharmacology - Florianopolis - SC ~Brazil. **F. Vanz:** None. **M.A. Bicca:** None. **M. Giachero:** None. **L.J. Bertoglio:** A. Employment/Salary (full or part-time):; Federal University of Santa Catarina - Department of Pharmacology - Florianopolis - SC ~Brazil.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.07/TT44

Topic: H.01. Animal Cognition and Behavior

Title: Selective PKR inhibition restores short-term memory deficits in apoE4-carrying mouse

Authors: D. IBGHI[1], *V. TAUPIN[2,1], P. BERNARDELLI[2], N. MOINDROT[1], P. GONIOT[1], E. GENET[1], C. VINCENT[1], V. ROUDIERES[1], V. FLEURY[1], A. KRICK[2], M. LOPEZ-GRANCHA[1]
[1]Neurodegeneration Research, Neurosci. Therapeut. Area, Sanofi, Inc., Chilly-Mazarin, France; [2]Sanofi R&D, CHILLY-MAZARIN, France

Abstract: Alzheimer's disease (AD) pathogenic mechanisms such as high brain levels of Aβ species, neuroinflammation, or presence of the main risk-promoting allele apoE4 lead to activation of the protein kinase R (PKR). PKR is a pro-apoptotic kinase that is activated by auto-phosphorylation, leading to phosphorylation of the translation initiation factor eiF2α. Phosphorylated eiF2α (peiF2α) selectively stimulates the translation of ATF4, a repressor of CREB-mediated memory function.
In AD patients, activation of PKR was evidenced by elevated levels of both phosphorylated PKR (pPKR) and peiF2α in brain. Increased pPKR was confirmed in CSF from AD individuals and it has been associated to faster cognitive decline.
In apoE4 carriers, enhanced pPKR levels have been seen in lymphocytes. Also, the presence of apoE4 has been associated with overexpression of ATF4 in brain. In agreement with these findings, mice carrying the human apoE4 isoform show both cognitive impairment and increased levels of pPKR and peiF2α in brain, when compared to apoE3 mice.
Here, we show that subchronic oral treatment with a brain blood barrier permeable selective PKR inhibitor (SAR439883) restores short-term memory deficits in apoE4 mice. This pro-cognitive effect was associated with highly selective, robust and dose-dependent inhibition of

PKR activity in the brain, as measured by both the PKR occupancy and the reduction of peIF2α levels. Remarkably, these effects were achieved from the lowest tested dose of SAR439883 with free brain concentration of 0.1 µM corresponding to 34 % PKR occupancy, which was consistent with the efficacy seen in vitro.
Our results indicate that PKR overactivity is strongly involved in the apoE4-mediated short-term memory deficits in mice. Moreover, they suggest that PKR inhibition could be a potential strategy for the treatment of diseases leading to memory impairment.

Disclosures: D. Ibghi: A. Employment/Salary (full or part-time):; Neurodegeneration Research, Neuroscience Therapeutic Area, Sanofi, Inc., Chilly Mazarin, France. **V. Taupin:** A. Employment/Salary (full or part-time):; Neurodegeneration Research, Neuroscience Therapeutic Area, Sanofi, Inc. **P. Bernardelli:** A. Employment/Salary (full or part-time):; sanofi R&D. **N. Moindrot:** A. Employment/Salary (full or part-time):; Neurodegeneration Research, Neuroscience Therapeutic Area, Sanofi, Inc. **P. goniot:** A. Employment/Salary (full or part-time):; Neurodegeneration Research, Neuroscience Therapeutic Area, Sanofi, Inc. **E. genet:** A. Employment/Salary (full or part-time):; Neurodegeneration Research, Neuroscience Therapeutic Area, Sanofi, Inc. **C. vincent:** A. Employment/Salary (full or part-time):; Neurodegeneration Research, Neuroscience Therapeutic Area, Sanofi, Inc. **V. roudieres:** A. Employment/Salary (full or part-time):; Neurodegeneration Research, Neuroscience Therapeutic Area, Sanofi, Inc. **V. fleury:** A. Employment/Salary (full or part-time):; Neurodegeneration Research, Neuroscience Therapeutic Area, Sanofi, Inc. **A. krick:** A. Employment/Salary (full or part-time):; sanofi R&D. **M. Lopez-Grancha:** A. Employment/Salary (full or part-time):; Neurodegeneration Research, Neuroscience Therapeutic Area, Sanofi, Inc..

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.08/TT45

Topic: H.01. Animal Cognition and Behavior

Support: UM-Flint Office of Research Award

Title: Cognitive responses to recurrent administration of intraperitoneal lipopolysaccharide

Authors: *B. J. KUPFERSCHMID[1], P. J. ROWSEY[2]
[1]Univ. of Michigan-Flint, Flint, MI; [2]The Univ. of North Carolina at Greensboro, Greensboro, NC

Abstract: Older individuals are susceptible to infections and recurrent infections are not uncommon. While an infection in older individuals may result in a decline in cognitive functioning, minimal literature exists on the impact of recurrent infections on cognitive

responses. In animals, symptoms of infections such as anorexia and cognitive changes, can be mimicked by administration of Lipopolysaccharide (LPS), a component of the cell wall of gram-negative bacteria. To model a recurrent infection, we examined the effects of intraperitoneal administration of 300 μg/kg LPS (n=5) or saline (n=5) for three consecutive days, followed by two weeks of rest and three additional days of LPS/saline injections. Aged (22 months) male Brown-Norway rats were trained 5 hours after injections for 6 days in the Morris water maze (MWM), a hippocampal-dependent test of spatial learning. Animals were assessed on days five and six of each week. Outcomes measured were spatial learning, food intake, and weight. Directional heading error (DHE), the degree of error in heading toward a hidden platform, and swim time latency were measured as indicators of spatial learning. Probe trials were conducted to assess retention. To determine if outcomes varied between the weeks, all outcomes were analyzed using week as a binary variable. LPS treated animals consumed less food ($p = .003$) and lost more weight than controls ($p = .001$). LPS treated animals also exhibited greater DHE over both weeks ($p = .062$) and while not significant, the LPS group took longer to reach the platform. Although there was not an experimental effect for probe trials, the experimental group travelled less in the correct quadrant. While there were no differences in treatment effect on spatial learning indicators or weight change between the two weeks, there were significant differences in treatment effect on mean food intake ($p < .0001$) with the experimental group increasing intake during week two. Despite some improvement in sickness responses in aged rats, as evidenced by food intake, recurrent exposure to LPS resulted in a trend toward spatial learning deficits including impaired directional heading. These results suggest that older individuals may be more at risk for cognitive impairment following recurrent infections.

Disclosures: B.J. Kupferschmid: None. **P.J. Rowsey:** None.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.09/TT46

Topic: H.01. Animal Cognition and Behavior

Title: Acute exposure to acetaminophen impairs object recognition memory in mice

Authors: *T. M. MILEWSKI[1], S. PATEL[1], C. A. STAPF[1], S. N. WIERBOWSKI[1], P. T. ORR[1,2]
[1]Neurosci. Program, [2]Psychology Dept., Univ. of Scranton, Scranton, PA

Abstract: The over the counter drug acetaminophen has been used for decades, but recent reports suggest that it has significant effects on cognition. Acute exposure to acetaminophen decreases emotional pain (Dewall et al, 2010) and dulls emotional processing (Durso, Lutterell &Way, 2015) in humans. However, it is not currently known if such acute exposure affects

memory. In this study, we show a dose-dependent impairment on hippocampal memory formation in an object recognition task in mice. 40 male mice were injected with vehicle, 10mg/kg, 50mg/kg, or 100mg/kg of acetaminophen immediately following training in a hippocampal-dependent object recognition memory task. Mice receiving 10 mg/kg of acetaminophen showed a significant preference for the novel object ($t(9) = 2.308$, $p = .046$), demonstrating intact memory for the familiar object, whereas there was not a significant novelty preference in the mice receiving 50 mg/kg ($t(9) = 1.618$, $p = .14$) or 100 mg/kg ($t(9) = .24$, $p = .816$) of acetaminophen. Subsequent to behavioral testing, brain samples were taken for analysis of cell-signaling events downstream of acetaminophen injection. Overall, these data demonstrate that acute, post-training exposure to 50 mg/kg or 100 mg/kg acetaminophen is sufficient to disrupt hippocampal-dependent memory consolidation.

Disclosures: T.M. Milewski: None. **S. Patel:** None. **C.A. Stapf:** None. **S.N. Wierbowski:** None. **P.T. Orr:** None.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.10/TT47

Topic: H.01. Animal Cognition and Behavior

Title: Microanatomy of fear memory consolidation and extinction within sub-regions of the prefrontal cortex and amygdala revealed by Arc and pERK/MAPK activity

Authors: *A. JACQUES[1,3,4], N. CHAAYA[1,3,4], A. BATTLE[2,3,4], L. R. JOHNSON[1,3,4,5]
[1]Sch. of Psychology and Counselling, Queensland Univ. of Technol., Brisbane, Australia; [2]Biomed. Sci., Queensland Univ. of Technol., Brisbane Adelaide Street, Australia; [3]Inst. of Hlth. and Biomed. Innovation, Queensland Univ. of Technol., Woolloongabba, Australia; [4]Translational Res. Institute, Queensland Univ. of Technol., Woolloongabba, Australia; [5]Ctr. for the Study of Traumatic Stress, Dept. of Psychiatry, USU Sch. of Med., Bethesda, MD

Abstract: Post-traumatic stress disorder is a memory-based disorder characterized by enhanced responses to perceived threats and a reduction in the ability to extinguish fearful memories. The generation of fearful or threat memories involve aspects of Pavlovian association between unconditioned, aversive (US) and conditioned, neutral (CS) stimuli. In order to understand the micro-anatomical organization of fear / threat memory processes and guide the development of brain region focused treatments for PTSD, a complete understanding of the micro-anatomical and molecular events underlying threat memory consolidation and extinction is required. Following Pavlovian fear conditioning (CS+USx3), we performed an ‘extended extinction’; rats received 3 days (d) of 20xCS, then 3d home cage prior to a fear memory test (3xCS). Phospho-ERK/MAPK (pMAPK) and Arc protein (Arc/Arg3.1) are sequentially activated markers of

neuroplasticity in response to memory. We investigated Arc and pMAPK immunoreactivity to measure micro-circuit localization of memory in the basolateral amygdala (BLA) known for its role in extinction and the infralimbic cortex (IL) reported to modulate the formation of new extinction memories, in fear memory and 'extended extinction' recall conditions. The relative contribution of pMAPK and Arc were measured in left and right hemispheres respectively (n = 40 animals). Initial results indicate numerically similar contributions of pMAPK and Arc activity in neurons occurring during extinction memory recall within the BLA and IL. Initial investigation also reveals specific sub regions and cortical layers differ in expression of pMAPK and Arc during extinction memory recall. These findings begin to identify the spatial and temporal properties of extinction memory micro-circuits and increase understanding of the mechanisms of pathological fear.

Disclosures: **A. Jacques:** None. **N. Chaaya:** None. **A. Battle:** None. **L.R. Johnson:** None.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.11/TT48

Topic: H.01. Animal Cognition and Behavior

Title: Hookworm infection attenuates hippocampus-dependent learning by inhibiting memory consolidation

Authors: *Z. KOLO[1], T. M. BROMBACHER[1], K. S. DE GOUVEIA[1], L. CRUYWAGEN[1], F. BOOLEY[1], M. DARBY[1], F. BROMBACHER[2]
[1]Pathology Dept., [2]Intl. Ctr. for Genet. Engin. and Biotech. (ICGEB) and Pathology Dept., Univ. of Cape Town, Cape Town, South Africa

Abstract: Previous studies have demonstrated the importance of IL-4 producing T cells in the meninges on cognitive function following hippocampal-dependent spatial learning and memory using Morris water maze task. This study aims to investigate the role of neuro-immune interactions on spatial learning and memory following infection with N.brasiliensis, a model of the hookworm N. Americanus. Cognitive function was investigated in mice using the Morris water maze. Mice were given 4 trials a day for 4 consecutive days and trained to locate a circular platform placed approximately 0.5 cm below water level in a circular Morris water maze. On tests of anxiety, thigmotaxis, percent immobility and immobility frequency were increased, while movement was significantly reduced in response to training.Distance swam, swim speed, and latencies to platform during acquisition were similar across groups, with fewer platform crossings by infected mice compared to naive during reference memory. These results correlated directly to reduced brain derived neurotrophic factor (BDNF) with infection, increased macrophages with training and infection, as well as increased microglia with training, but not

infection. Both speed and distance swam were ruled out as factors for any cognitive differences in this study. Because infection plays a role in anxiety defining behavior, we conclude that anxiety interferes with memory consolidation during the acquisition phase of the task as characterized by impaired reference memory, and reduced BDNF. We also conclude that an increase in macrophages due to infection inhibits learning whereas an increase due to training enhances learning. However, the presence of macrophages during infection may also play a role in enhanced learning. Infection alone dampened production of IL-4 by macrophages, while infection with training, showed increased IL-4 production. Overall, we conclude that hookworm infection is detrimental to learning and memory by means of inducing anxiety that interferes with memory consolidation, and that macrophages recruited during infection impair reference memory possibly by production of both IL-4 and IL-13 that in above baseline levels affect memory consolidation without "harming" acquisition. In addition to showing the involvement of microglia in cognitive function we also show that their numbers are reduced during hookworm infection. We show that IL-13, but not necessarily IL-4 appears to steer microglia driven learning and memory in the absence of infection.

Disclosures: Z. Kolo: None. **T.M. Brombacher:** None. **K.S. De Gouveia:** None. **L. Cruywagen:** None. **F. Booley:** None. **M. Darby:** None. **F. Brombacher:** Other; International Centre for Genetic Engineering and Biotechnology.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.12/TT49

Topic: H.01. Animal Cognition and Behavior

Support: 5T32MH019524-23

R01-MH100822

Title: High demand of hippocampal glucose and lactate during memory formation in early development

Authors: *E. CRUZ[1], A. TRAVAGLIA[2], C. M. ALBERINI[3]
[1]Ctr. for Neural Sci., New York Univ. Ctr. for Neural Sci., New York, NY; [3]Ctr. for Neural Sci., [2]New York Univ., New York, NY

Abstract: Hippocampal glycogenolysis and astrocyte-neuronal lactate metabolic coupling are necessary in the adult rat hippocampus for the consolidation of inhibitory avoidance (IA) memory as well as for in-vivo hippocampal long-term potentiation. Here, we investigated the role of hippocampal glucose and lactate metabolism in memory consolidation during early

development. Specifically, we focused on rats at postnatal day 24 (PN24), a young age when rats have just acquired the ability to form strong and long lasting hippocampal-dependent memories. Quantitative polymerase chain reaction array (qPCR-array) of dorsal hippocampal RNAs revealed that 40 out of 84 glucose metabolism-related genes were up-regulated in PN24 rats compared to the adult rats (PN80). In addition, IA training led to significant increases in both hippocampal extracellular glucose and expression of the glucose transporters (GLUT1 and GLUT3) in PN24 rats. Inhibition of hippocampal glycogenolysis with 4-dideoxy-1, 4-imino-D-arabinitol (DAB), or knockdown of GLUT3 or lactate transporters (Monocarboxylate transporters 1, 2, and 4) impaired IA memory formation in PN24 rats. Unlike in adult rats, both, D-glucose and L-lactate persistently rescued these memory impairments. Furthermore, higher levels of D-glucose and L-lactate were required to rescue the memory impairments of PN24 rats compared to adults, suggesting that the developing hippocampus requires higher levels of these metabolic substrates to form long-lasting memories. We conclude that hippocampal metabolic requirements and possibly mechanisms underlying memory formation during development differ from those of adults.

Disclosures: **E. Cruz:** None. **A. Travaglia:** None. **C.M. Alberini:** None.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.13/TT50

Topic: H.01. Animal Cognition and Behavior

Support: NIH Grant AA019462

NIH Grant 2T32AA014127-11

Title: Alcohol exposure during gestational development alters spatial extinction behavior in adult male and female rats

Authors: *C. M. MAGCALAS[1], J. WAGNER[2], D. D. SAVAGE[2], D. A. HAMILTON[1]
[1]Psychology, [2]Neurosciences, Univ. of New Mexico, Albuquerque, NM

Abstract: Prenatal alcohol exposure (PAE) is associated with structural and physiological changes that impact the central nervous system and can result in persistent negative consequences in a broad spectrum of cognitive and behavioral domains including deficits in motor behavior, social behavior, and behavioral flexibility. Previous studies have characterized the influence of PAE on spatial navigation acquisition and extinction through various behavioral paradigms including the Morris water task (MWT). The current study focuses on examining the behavioral consequences of PAE on spatial extinction behavior through the use of the MWT.

Pregnant rat dams voluntarily consumed saccharin (SAC) water containing 0% or 5% ethanol (EtOH) for 4 hours per day during the entire gestational period. Male and female pups matured and were tested in the MWT in adulthood (>90 days old). In order to assess extinction behavior the animals were tested in a 5-day hidden platform protocol. Days 1-4 of the hidden protocol consisted of 12 training trials. At the end of day 4, the animals were tested in 3 consecutive no-platform probe trials to assess extinction behavior. Day 5 began with one no-platform probe trial to assess spontaneous recovery followed by 12 retraining trials. All of the animals successfully learned the hidden platform goal location during the initial training period. All of the male animals (PAE and controls) failed to extinguish, which was evident by the consistent short latency to reach the learned target location. The control females successfully extinguished, but the ethanol exposed females failed to extinguish the learned behavior. These outcomes suggest that animals exposed to moderate levels of ethanol during gestation have intact spatial acquisition abilities, but may have distinct extinction behaviors that may be sex specific and can be influenced by PAE. [Supported by grant AA019462 to DH].

Disclosures: C.M. Magcalas: None. **J. Wagner:** None. **D.D. Savage:** None. **D.A. Hamilton:** None.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.14/TT51

Topic: H.01. Animal Cognition and Behavior

Support: Natural Sciences and Engineering Research Council of Canada (NSERC) Discovery Grant #40352

Campus Alberta for Innovation Program Chair, Alberta Alzheimer Research Program

Canadian Institute for Advanced Research grant

Title: How much gestational noise stress impairs reproduction activates HPA axis and damages behavior in mice

Authors: *Z. JAFARI, J. MEHLA, B. E. KOLB, M. H. MOHAJERANI
Dept. of Neurosci., Univ. of Lethbridge, CCBN, Lethbridge, AB, Canada

Abstract: Maternal stress is a common adversity during pregnancy. Noise exposure is a known environmental pollutant whose adverse effect on health has been less studied. We used “noise” as a gestational stress compared with physical stress to explore the effect of stress during gestation on hypothalamic-pituitary-adrenal (HPA) axis activation, cognitive performance, and behavior in the dams and their offspring. Noise stress caused higher rates of resorbed embryos

and reduction of litter, as well as an increase in anxiety-like behavior and reduced time spent exploring new environment compared to the physical stress and control groups. It also severely impaired the HPA axis activation, as well as cognitive, memory, and balance function as large as or stronger than the physical stress in both mothers and offspring. Both prenatal stresses also caused a strong susceptibility to environmental experiences as stressful conditions in the offspring. The findings suggest the significance of conservation against loud noise exposure in daily living, as well as need to further notice to the different aspects of gestational stress on health.

Key Words: Noise stress, uterus receptivity, reproduction, HPA axis, behaviour, anxiety

Disclosures: Z. Jafari: None. **J. Mehla:** None. **B.E. Kolb:** None. **M.H. Mohajerani:** None.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.15/TT52

Topic: H.01. Animal Cognition and Behavior

Title: Acute restraint stress impairs object recognition memory in female mice

Authors: C. A. STAPF[1], K. N. WALTER[1], K. E. TALERICO[1], *P. T. ORR[2,1]
[1]Neurosci. Program, [2]Psychology Dept., Univ. of Scranton, Scranton, PA

Abstract: It has been well established that hippocampal-dependent memory formation in rodents is affected by acute and chronic stress. However, this has been established primarily in male rodents, even though hippocampal-dependent tasks, such as object recognition, are sensitive to steroid hormones such as estradiol and progesterone, which cycle in intact females. Given the recent decision by the National Institutes of Health to highlight sex as a biological variable in biomedical research, it is critically important to establish baseline effects such as this in females. In this study, we examined intact female mice to determine if acute restraint stress impaired performance on a novel object recognition memory task, as it does in male mice. Adult (8 week old) intact female mice were trained in an object recognition task and subsequently restrained for 1 hour. During testing, control (unrestrained) mice showed a significant preference for the novel object ($t(11)=2.368$, $p=0.037$) suggesting intact memory. Restrained mice did not show a preference for the novel object ($t(12)= -.189$, $p=0.854$), suggesting a lack of memory. Two weeks after behavioral testing, mice were again restrained for one hour and hippocampus was dissected out for analysis of cell-signaling cascades. Acute restraint stress significantly impairs hippocampal-dependent consolidation of object recognition memory in intact female mice.

Disclosures: C.A. Stapf: None. **K.N. Walter:** None. **K.E. Talerico:** None. **P.T. Orr:** None.

Poster

424. Memory Consolidation and Reconsolidation: From Epigenetics to Gestation

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 424.16/TT53

Topic: H.01. Animal Cognition and Behavior

Title: Phases of the estrous cycle modulate renewal of appetitive and fear behavior

Authors: *E. HILZ, M. MONFILS, H. LEE
Psychology, Univ. of Texas At Austin, Austin, TX

Abstract: The gonadal hormone estradiol has been shown to increase synaptic plasticity in the hippocampus, an area of the brain known to play an important role in contextual information processing. The hippocampus mediates both renewal and memory retrieval of conditioned fear after extinction, but little research has examined if natural fluctuations in estradiol across the estrus cycle modulate the expression of contextual learning and memory. In the present experiment, we examine whether the level of estradiol present during extinction - either low-estradiol during the metestrus/diestrus (M/D) phases of the estrous cycle or high-estradiol during the proestrus (P) phase of the estrous cycle - plays a role in contextual renewal of appetitive and fear behavior. Female and male Sprague-Dawley rats were exposed to both appetitive and fear acquisition and extinction procedures and tested for renewal of conditioned behavior in an ABBA design (i.e., acquisition in context A, extinction in context B, and test in both contexts A and B). Appetitive results suggest that if female rats were in the P phase during extinction, they show significantly higher renewal of behavior in the original training context as compared to the females that were in M/D phase during extinction. Male rats did not significantly differ from either group. Preliminary results suggest that the estrous cycle may modulate renewal of extinguished fear differently. In general, our results suggest that estradiol levels might modulate contextual information processing during extinction.

Disclosures: E. Hilz: A. Employment/Salary (full or part-time):; University of Texas at Austin. **M. Monfils:** A. Employment/Salary (full or part-time):; University of Texas at Austin. **H. Lee:** A. Employment/Salary (full or part-time):; University of Texas at Austin.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.01/TT54

Topic: H.01. Animal Cognition and Behavior

Support: NIH RO1 MH110300

Whitehall Foundation Research Grant 2015-12-71

BBRF NARSAD 23017

Title: Anterior thalamus regulates interactions between hippocampus and retrosplenial cortex

Authors: *N. A. KAMBI, J. M. PHILLIPS, Y. B. SAALMANN
Dept. of Psychology, Univ. of Wisconsin-Madison, Madison, WI

Abstract: Episodic memory is the capacity allowing us to consciously remember objects and events from our personal past. A large body of evidence suggests that the hippocampus and retrosplenial cortex play an important role in episodic memory. However, the nature of interactions and the information flow between these brain regions for supporting episodic memory and spatial processing remains unclear. The anterior thalamus is reciprocally connected with both the hippocampus and retrosplenial cortex, and anterior thalamic lesions can give rise to memory deficits. We thus hypothesized that the anterior thalamus supports episodic memory by regulating information transmission between the hippocampus and retrosplenial cortex. To test our hypothesis, we simultaneously recorded spikes and local field potentials (LFPs) from the anterior thalamus, hippocampus (subiculum) and retrosplenial cortex of two macaques performing a visuospatial episodic-like memory task. We used diffusion MRI to target linear electrode arrays to interconnected sites of this hippocampo-thalamo-cortical network (1mm isotropic, 60 diffusion directions, b=1000s/mm^2 and NEX=14, using a 3T GE MR750 scanner and 16-channel receive-only head coil), and structural MRI of electrodes in situ to confirm electrode positions (0.5mm isotropic). Our preliminary electrophysiological data show bidirectional conditional Granger causal influences in the 5-15Hz range between the hippocampus and retrosplenial cortex during memory retrieval. During this time, neurons in the anterior thalamus (ventral division) responded preferentially to particular learnt visual scenes. We also measured increased coherence between thalamic spikes and the LFPs in both the hippocampus and retrosplenial cortex, in the 5-15Hz range. Further, the anterior thalamus showed increased conditional Granger causal influences on the hippocampus and retrosplenial cortex in the same frequency range. These preliminary data suggest that the anterior thalamus regulates information transmission between the hippocampus and retrosplenial cortex based on episodic memory demands. The underlying mechanisms appear to include the anterior thalamus synchronizing activity between the hippocampus and retrosplenial cortex. Overall, this study supports an important role for the anterior thalamus in the extended hippocampal system supporting episodic memory.

Disclosures: **N.A. Kambi:** None. **J.M. Phillips:** None. **Y.B. Saalmann:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.02/TT55

Topic: H.01. Animal Cognition and Behavior

Support: NIMH R01MH108729

Title: Neuronal activity in the retrosplenial cortex of rats performing a visuospatial attention task

Authors: *E. HWANG[1], F.-C. YANG[3], T. K. JACOBSON[1], R. D. BURWELL[2]
[1]CLPS, [2]Dept. of Cognitive, Linguistic, and Psychological Sci., Brown Univ., Providence, RI;
[3]Dept. of Psychology, Univ. of Michigan, Ann Arbor, MI

Abstract: Research in primates and rodents has addressed the role of the retrosplenial cortex (RSC) in a variety of spatial functions including navigation, contextual learning, and the integration of information across spatial reference frames. Anatomical studies of the rodent brain show that RSC is connected with other regions that process spatial information for a variety of purposes, including the hippocampus (navigation and spatial memory), postrhinal cortex (POR, representation of context), and posterior parietal cortex (PPC, visuospatial attention and action selection). An open question is what *core function* of RSC could support the variety of spatial information processing observed in related brain regions. We hypothesize that RSC supports online integration of information across space, time, contexts, and reference frames for use by other brain regions for a variety of functions. To address this hypothesis, we recorded from the RSC in rats performing the visuospatial attention (VSA) task. This task is a purely visual task that was adapted from the five choice serial reaction time task (Bari et al, 2008) for a double-sided, bowtie shaped enclosure atop the Floor Projection Maze (Jacobson et al., 2014). The task involves top-down and bottom-up visuospatial attention, multiple spatial reference frames, and the processing of reward. The animals are trained to attend to three locations and to approach the spot that is briefly illuminated (see figure). Trials alternate east to west. Approach to the correct location is followed by reward. Task epochs included pre- and post-stimulus, pre- and post-selection, and reward. We found that the majority of recorded RSC cells signal both allocentric and egocentric location and also integrate outcome with multiple task events. Additionally, RSC activity increases as the rat progresses through different task epochs, suggesting integration of information across task epochs. Analyses of neuronal correlates will be presented and patterns of selectivity in RSC will be compared with those of the PPC and POR recorded in rats performing the same VSA task.

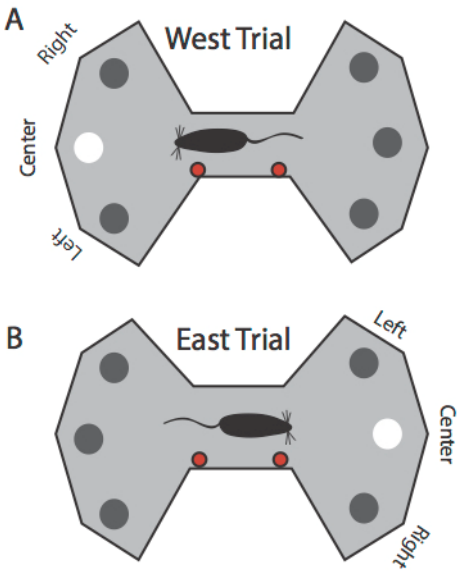


Fig 1. Shown above is an animal in ready position. A shows animal approaching correct stimulus on the West trial and B on the East trial. Animals are rewarded at the reward port (red dots).

Disclosures: **E. Hwang:** None. **F. Yang:** None. **T.K. Jacobson:** None. **R.D. Burwell:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.03/TT56

Topic: H.01. Animal Cognition and Behavior

Support: NSF GRFP 1058262 to VJE

NIMH R01MH108729 to RDB

Title: Object-location conjunctive coding in the parahippocampal network

Authors: *V. J. ESTELA[1], A. FAROVIK[2], R. D. BURWELL[2]
[1]Neurosci., [2]Cognitive Linguistic and Psychological Sci., Brown Univ., Providence, RI

Abstract: The formation of episodic memories requires a network capable of representing objects within a complex context, allowing for recollection of events in time and space. These contexts consist of items, places, patterns, as well as the specific spatial arrangement of these elements. Structures in the medial temporal lobe, specifically the perirhinal (PER) and postrhinal (POR) cortices, are crucial for contextual learning. Our overarching hypothesis is that the POR integrates object information from the PER with spatial information from the posterior parietal and retrosplenial cortices to represent the spatial layout of objects, patterns, and features in the local environmental context. Further, we hypothesize that the hippocampus (HC) relies on contextual representations from the POR for associative learning and episodic memory. Prior studies reported that both the POR and HC exhibit object-location conjunctive coding (Furtak, et al., 2012; Komorowski, et al., 2009). Such conjunctions could be considered a signature of context representations, but they could also reflect associative learning. Indeed, object-location conjunctions emerge with learning in the HC. The timing of emergence of object-location conjunctions in the POR is unknown. One possibility is that the object-location conjunctions in the POR reflect a complete representation of the local spatial environment, whereas the HC uses this representation of the spatial environment or context for episodic memory and in the formation of associative memories. If our hypothesis is correct, object-location conjunctions should be present in the POR before they emerge in the HC. To test this prediction, we will record in the POR and HC during performance on the location biconditional discrimination (locBCD) task in the floor projection maze. In this task, presentation of a pair of objects alternates between the east and west sides of a bowtie-shaped maze. The east-west location determines which of the two objects is correct. Finding object-location conjunctions in the POR prior to the HC would be consistent with the interpretation that object-location conjunctive coding in the HC relies on POR representations of context. Future studies will address this interpretation in circuit analysis experiments that utilize dual-site recording of the POR and HC as well as experiments that combine electrophysiology and optogenetics in rats performing the locBCD task.

Disclosures: **V.J. Estela:** None. **A. Farovik:** None. **R.D. Burwell:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.04/TT57

Topic: H.01. Animal Cognition and Behavior

Title: Bio-plausible models of ACC function in a novel probabilistic reversal learning task

Authors: *R. M. FRANCIS[1], R. A. WIRT[2], J. M. HYMAN[3]
[1]Psychology, UNLV, Las Vegas, NV; [3]Psychology, [2]Univ. of Nevada Las Vegas, Las Vegas, NV

Abstract: The anterior cingulate cortex (ACC) is often associated with learning and decision-making due in part to its vast array of behaviorally-related electrophysiological signals. Most notably, some ACC neurons carry signals that classify outcomes as either rewarding or non-rewarding, here called outcome-tracking signals. Additionally, experimental evidence suggests that the ACC generates the feedback-related negativity signal (FN), which is an event-related potential modulated by the probability of receiving different outcomes. Currently, the roles of ACC outcome-tracking signals and the FN in learning, decision-making, and ultimately preference formation are unknown. Here we report a novel behavioral task that helps to better characterize these processes. Our probabilistic reversal task allowed us to observe the formation of different behavioral preferences within rodent subjects multiple times in a single session. This task featured two ports, one with a 75% probability of yielding a reward, and the other with a payout probability of 25%. On *cued* trials, subjects were randomly prompted to nose-poke at one of the two ports. This action would trigger one of two feedback scents to be pumped directly into the port. One scent signaled that reward in the form of a sugar pellet would follow 1 second later, and the other scent signaled that no reward would be available on that trial. After every four *cued* trials, there was a *probe* trial where both ports would be active. *Probe* trial choices permitted us to assess the degree of behavioral preference for the 75% port and how preference was affected by recent *cued* trial outcomes. We set a behavioral preference criterion of three consecutive *probe* trial choices of the 75% port ($p<.05$). Once this threshold was met, the reward probabilities of the two ports were reversed, the threshold counter was wiped, and the task continued. Some subjects achieved up to 5 reversals in a single 1-hour session. We created a series of computational models to characterize behavioral transitions between preferences that existed before and after reversals. Some models used mechanisms based on ACC outcome-tracking signals and the FN to assess whether these models outperformed conventional models' ability to fit the data. Analyses revealed that the bio-plausible models (featuring outcome-tracking signals and FN homologues) more accurately predicted when behavioral preferences formed in comparison to their conventional versions, suggesting that these outcome-related ACC signals have a specific relationship, and actually play an integral role in decision-making and preference formation.

Disclosures: **R.M. Francis:** None. **R.A. Wirt:** None. **J.M. Hyman:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.05/TT58

Topic: H.01. Animal Cognition and Behavior

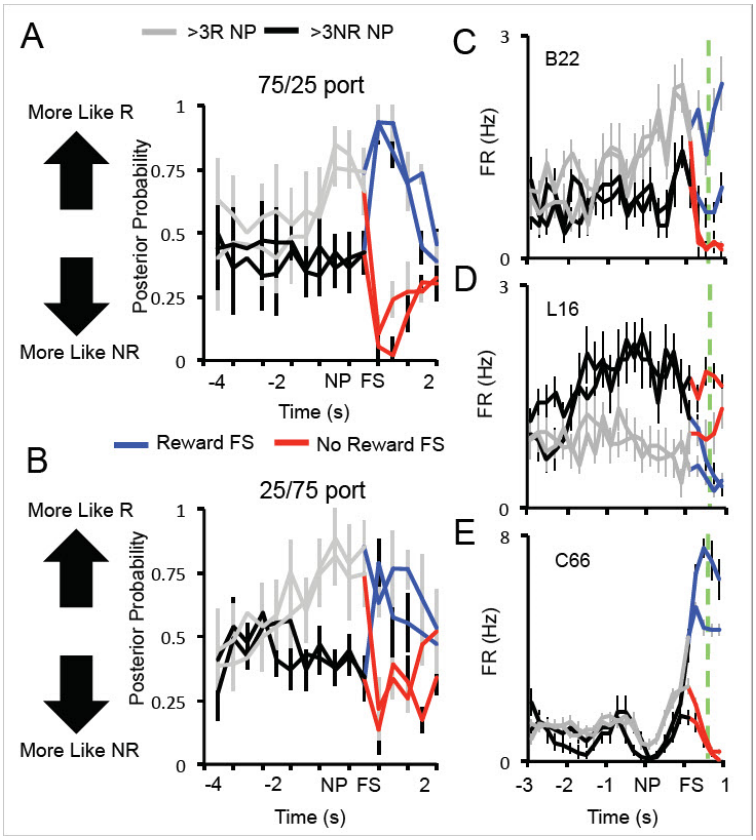
Support: MOP-93784

MOP-84319

Title: Expectation and outcome representations combine to create a novel signal in ACC neurons underlying the FN

Authors: *J. M. HYMAN[1], C. B. HOLROYD[2], J. K. SEAMANS[3]
[1]Psychology, Univ. of Nevada Las Vegas, Las Vegas, NV; [2]Univ. of Victoria, Victoria, BC, Canada; [3]Psychiatry, UBC, Vancouver, BC, Canada

Abstract: The function of the anterior cingulate cortex (ACC) remains controversial, yet many theories have emphasized its importance in dynamically changing environments where expectations are frequently violated. This view is motivated in part by observations of a human event-related potential termed the feedback-related negativity (FN) that is evoked over the ACC whenever such violations occur. Although the FN is thought to depend on prediction errors, it is not clear how predictions and prediction errors are encoded in ACC and how they lead to an FN. To address these issues we recorded from ACC neurons while rats performed a task identical to one that reliably evokes a robust FN in humans. On each trial the rats were cued to perform a nose poke (NP) at one of 3 ports (each with a different payout probability) at which point a feedback scent was delivered informing the rat whether the trial would be rewarded or not. A subset of ACC neurons was found that encoded expected outcomes according to recent outcome history. Specifically, responses during the NP mimicked responses to either the positive or negative feedback scents on trials preceded by many rewarded or non-rewarded outcomes respectively. The representation of expected outcomes persisted throughout the trial until the actual outcome feedback scent was delivered, at which point the ensembles shifted from representing the expected to actual outcome on incongruent trials. This shift occurred at the same point in the trial where an FN-like response was observed in ACC field potentials (Warren et al., 2015). Thus the present study identified the requisite neural components of prediction error signaling in ACC and demonstrated the novel way in which they interact to produce an FN.



Disclosures: **J.M. Hyman:** None. **C.B. Holroyd:** None. **J.K. Seamans:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.06/TT59

Topic: H.01. Animal Cognition and Behavior

Title: It's about time: Electrophysiological evidence for temporally mediated consolidation of spatial memories

Authors: *R. A. WIRT[1], L. A. CREW[1], K. R. ZHA[1], N. L. KAPLAN[1], R. M. FRANCIS[1], J. M. HYMAN[2]
[2]Psychology, [1]Univ. of Nevada Las Vegas, Las Vegas, NV

Abstract: Formation and retrieval of new spatial information is highly dependent on the hippocampus (HC), but over time these memories become increasingly reliant on other areas, including the anterior cingulate cortex (ACC) (Bontempi et al., 1999). We hypothesized that if this were the case then the retrieval of older spatial memories would engender different

interactions between the HC and ACC than more recent memories would. Specifically, there should be changes in synchrony between these two areas and the direction of the interactions should shift from the HC leading for recent memories to the ACC leading for more remote retrieval. To assess this, we recorded both single units and local field potentials from the ACC and HC as animals were exposed to unique spatial environments and then re-exposed to those same environments at differing time delays (1-14 days). Behavioral data revealed a significant decrease in exploration-related movement as early as the second exposure regardless of the delay period, indicating that the animals quickly became familiar with the environments. We found that neural synchrony between the ACC and HC was significantly stronger on the remote retrieval days, despite no discernible behavioral differences from recent retrieval days. These effects were found in theta and gamma band coherence, phase synchrony, and ACC unit phase locking to HC theta. Time-lag correlation analysis revealed that while on days 1-7 HC theta activity led the ACC, on day 14 this effect reversed and theta in the ACC now led the HC. Additionally, there were no significant changes in ACC-HC synchrony until 14-days after initial exposure regardless of the number of previous experiences, supporting the conclusion that the operative variable affecting these interactions was the passage of time. We also observed increases in bilateral ACC synchrony on day 14, which may be indicative of an ACC mediated retrieval of remote memories. These results reveal a clear electrophysiological signature of spatial memory consolidation to the ACC, and they pinpoint that the observed changes were due to time passing and not other possible factors.

Disclosures: **R.A. Wirt:** None. **L.A. Crew:** None. **K.R. Zha:** None. **N.L. Kaplan:** None. **R.M. Francis:** None. **J.M. Hyman:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.07/TT60

Topic: H.01. Animal Cognition and Behavior

Support: DFG: SonderForschungBereich (SFB) 874

Mercator Stiftung

Title: Familiarity-induced activity patterns in the LEC and PER in the absence of a functional hippocampus

Authors: *L. MAHNKE[1,2], E. ATUCHA[1,2], T. KITSUKAWA[3], M. SAUVAGE[1,2,4,5]
[1]Functional Architecture of Memory Dpt, Leibniz Inst. For Neurobio., Magdeburg, Germany; [2]Functional Architecture of Memory Unit, Mercator Res. Group, Ruhr-University Bochum,

Germany; [3]Osaka Univ., Suita, Japan; [4]Med. Faculty, Functional Neuroplasticity Dept, [5]Ctr. for Behavioral Brain Sci., Otto von Guericke Univ., Magdeburg, Germany

Abstract: Recognition memory relies on the recollection and the familiarity processes. It is a consensus that the hippocampus supports recollection, but a major debate remains whether this brain area also supports familiarity or whether the parahippocampal region does, especially the perirhinal (PER) and the lateral entorhinal (LEC) cortices. Using a high resolution molecular imaging technique based on the detection of the immediate-early gene *Arc*, closely tight to synaptic plasticity, memory performance and commonly used to map activity in the medial temporal lobe, we recently showed that the PER and the LEC are selectively recruited during familiarity-only judgements in rats with intact hippocampal function, while the hippocampal subfields CA1 and CA3 are not (see Atucha et al., *submitted*). However, it remained unclear whether this recruitment was completely independent of the hippocampus. To address this question, we imaged brain activity in the PER and the LEC of rats with lesioned hippocampus performing a delay non-match to sample memory task, as performance under these conditions rely on familiarity (Fortin et al., *Nature*, 2004), and investigated whether activity patterns in the PER and the LEC were affected when hippocampal function was compromised. Preliminary data indicate this is not the case, suggesting that familiarity-induced patterns of activity detected in the LEC and the PER are independent of the hippocampus, and giving further support to the claim that the PER and the LEC support familiarity and not the hippocampus.

Disclosures: L. Mahnke: None. **E. Atucha:** None. **T. Kitsukawa:** None. **M. Sauvage:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.08/DP11/TT61 (Dynamic Poster)

Topic: H.01. Animal Cognition and Behavior

Support: Howard Hughes Medical Institute

5U01NS094296 (BRAIN Initiative)

MURI, W911NF-12-1-0594

Kavli Foundation

NSF IGERT

NSF GRFP

Simons Collaboration on the Global Brain

Title: Identifying olfactory neural circuits in adult *Drosophila* using high-speed whole brain functional imaging (SCAPE)

Authors: *N. MISHRA[1], W. LI[2], E. S. SCHAFFER[1], V. VOLETI[2], E. M. HILLMAN[2], R. AXEL[1]
[1]Neurobio. and Behavior, [2]Biomed. Engin., Columbia Univ., New York, NY

Abstract: While imaging small brain regions or even single neurons has revealed a lot about neural processing, the insights we gain from looking at larger populations are very different. Imaging neurons across the brain affords us the possibility to observe the relationship of activity in one part of the brain versus another. Toward this goal, we are using high-speed light sheet microscopy to do pan-neuronal calcium imaging in adult *Drosophila*.
We use Swept Confocally Aligned Planar Excitation (SCAPE) microscopy to observe the activity at single-cell resolution across the entire brain volume at speeds up to 20 volumes per second. Due to SCAPE's unique optical set up, a form of light-sheet imaging that uses a single objective lens and a novel scanning-descanning approach, we are able to do high-speed volumetric imaging in a non-transparent animal. During our in vivo imaging, flies expressing pan-neuronal GCaMP are subjected to an odor stimulus controlled via an olfactometer while performing tracked locomotive behavior on an air-supported ball. We test the performance of SCAPE by seeing activity distributed across the brain as our flies navigate a virtual environment. The spatiotemporal capabilities of this methodology will allow us to subsequently study the neural dynamics of more complex behaviors and internal states.

Disclosures: N. Mishra: None. **W. Li:** None. **E.S. Schaffer:** None. **V. Voleti:** None. **E.M. Hillman:** None. **R. Axel:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.09/TT62

Topic: H.01. Animal Cognition and Behavior

Support: National Key Basic Research Program of China 2012CB966300, 2013CB530900

Natural Science Foundation of China 31371114, 31200852, 31171060

RGC T13-607/12R, C1014-15G, 561213M, 561313M, 11101215M, 11166316M

HMRF 03141196,01121906

Title: Cholecystokinin from entorhinal cortex switches long-term potentiation in hippocampus

Authors: *J. SU[1], W. YE[2], J. HE[1]
[1]City Univ. of Hong Kong, Hong Kong, China; [2]Guangzhou Inst. of Biomedicine and Hlth., Guangzhou, China

Abstract: Our earlier studies showed that activation of cholecystokinin (CCK) neurons that originated from the entorhinal cortex (EC) induces long-term potentiation (LTP) and neuroplasticity in the auditory cortex. Lesions in EC or hippocampus will lead to memory deficits which have received intensive studies. In the present study, we found CCK peptide knockout mice (CCK-KO) showed severe deficits in spatial memory and hippocampal CA3-CA1 LTP formation induced by high-frequency (HF) electrical stimulation (ES) in vivo. We also found the same HF ES stimulation still induced CA3-CA1 LTP in CCK B-receptor knockout mice (CCKBR-KO), which exhibited normal spatial memory, while CCK A-receptor antagonist Devazepide totally blocked the LTP formation. We thus hypothesize that CCK released in respond to HF ES stimulation participates in LTP formation, the effect of which is mediated by CCKAR in hippocampus CA3-CA1 pathway. We further injected Cre-dependent ChR2-eYFP virus into either hippocampus or EC of CCK-Cre mice, and applied light stimulation in hippocampus. While HF light stimulation of CCK positive neuron in hippocampus, followed by low frequency (LF) ES stimulation of CA3, did not induce LTP, the same HF light manipulation of CCK positive projections from entorhinal cortex initiated LTP in CA3-CA1 pathway. Increased CCK concentration was also detected in hippocampus after HF light stimulation of these EC-originated terminals. We explain that HF stimulation of the EC originated CCK terminals induced CCK release in hippocampus, and LF ES stimulation of CA3 could induce LTP in the presence of CCK. The enhanced connectivity within hippocampus induced by this HF light-stimulation protocol was further supported in a spatial fear memory test, in which the CCK-Cre mice showed fear against both the footshock-given region and the HF light-paired region.

Disclosures: **J. Su:** None. **W. Ye:** None. **J. He:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.10/TT63

Topic: H.01. Animal Cognition and Behavior

Support: 1R21AA024983-01A1

P50AA022534

Title: Impaired performance in an object place paired associate task by a rat model of moderate prenatal alcohol exposure

Authors: *L. M. SANCHEZ[1], J. K. GOSS[1], J. WAGNER[2], S. DAVIES[2], D. A. HAMILTON[1], D. D. SAVAGE II[2], B. J. CLARK[1]
[1]Psychology, Univ. of New Mexico, Albuquerque, NM; [2]Neurosciences, Univ. of New Mexico Sch. of Med., Albuquerque, NM

Abstract: Memory impairments, including spatial and object processing, are often observed in individuals with Fetal Alcohol Spectrum Disorder. Although there is currently no consensus regarding the neuroanatomy for these memory deficits, much attention has been directed towards the hippocampus, which displays significant alterations after prenatal alcohol exposure (PAE). The parahippocampal cortex, which forms a major input-output relationship with the hippocampus, has a role in processing information about spatial locations and environmental features such as landmarks and objects. In the present study, we tested a moderate PAE rat model (Savage et al., 2010) in an object-place paired associate (OPPA) task, which has previously been shown to require hippocampal and parahippocampal processing. In brief, the OPPA task was composed of training rats to discriminate between an identical pair of objects presented in 180° opposite arms of a radial arm maze. Animals were given a total of 10 trials per day over 14 days of training and were rewarded with a piece of cereal after the correct selection of an object. Thus, animals were required to select a particular object on the basis of spatial location. We observed that PAE rats (n = 16) made significantly more errors than saccharin control rats (n = 16) during acquisition of the OPPA task. In Experiment 2, animals were trained in a reversal task which required rats to once again discriminate between identical pairs of object presented in 180° opposing arms. Although the same objects were used as in Experiment 1, the opposite object was rewarded in each arm. Rats were therefore required to flexibly learn a new object-place paired association. Again, animals were given a total of 10 trials per day over a 14 day training schedule. In the reversal task, PAE and saccharin control rats performed comparably, however there were mean differences in acquisition performance on days 7 through 12. Following the reversal task, in which training is halted and restarted in 96 hours to test for memory, PAE rats showed a significant, but mild decrease in performance. In Experiment 3, rats performed an object discrimination task in which rats were trained in a single maze to accurately select a rewarded object from a pair of objects. In this task, moderate PAE and saccharin control rats exhibited comparable performance. The observations of impaired performance, slower learning, and lack of flexibility by moderate PAE animals is discussed in relation to theories of the hippocampal-parahippocampal and limbic system basis of object-place memory.

Disclosures: L.M. Sanchez: None. **J.K. Goss:** None. **J. Wagner:** None. **S. Davies:** None. **D.A. Hamilton:** None. **D.D. Savage II:** None. **B.J. Clark:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.11/TT64

Topic: H.01. Animal Cognition and Behavior

Support: NIGMS (P30GM103400),

NIAAA (P50AA022534)

NIAAA (R21AA024983)

Title: Reduced spatial coding of hippocampal place cells following moderate prenatal alcohol exposure

Authors: *R. E. HARVEY, J. K. GOSS, T. RIGG, L. E. BERKOWITZ, L. M. SANCHEZ, J. L. WAGNER, D. D. SAVAGE, D. A. HAMILTON, B. J. CLARK
Dept. of Psychology, Univ. of New Mexico, Albuquerque, NM

Abstract: A common behavioral manifestation of prenatal alcohol exposure (PAE) is the loss of spatial memory, which is marked by a general inability to recognize familiar places and to accurately navigate from one location to another. Spatial deficits in PAE have been linked to the hippocampus, which displays reduced synaptic plasticity in moderate PAE models. In vivo electrophysiological recordings from the hippocampus have shown that specific neurons called place cells within CA1, CA3, and dentate gyrus subregions fire when animals enter specific regions of the environment. Each hippocampal place cell fires in a different location indicating that a large population of these cells cover the spatial layout of the environment. It is currently unknown if hippocampal deficits observed PAE animals negatively impact place cell firing. Thus, we performed electrophysiological recordings from the hippocampi (CA1 and CA3) of moderate PAE adult rats (Savage et al., 2010) and saccharin control adult rats. Rats were implanted with high density tetrode arrays aimed at the CA1 and CA3 regions. Following implantation, rats were trained to complete laps on a narrow linear track across multiple 10 - 20 minute sessions while single unit and local field potentials were recorded. Around 200 hippocampal neurons were identified as place cells based on their spatial and firing characteristics. Our results show that hippocampal place cells from PAE rats have severe spatial coding impairments such as reduced spatial information content, increased sparsity, and expanded firing field widths which indicate a lack of place cell spatial specificity. Further, indicative of network deficiencies, PAE rats have reduced theta phase locking and theta power compared to saccharin control rats. In sum, the failure of hippocampal place cell firing to selectively discriminate between spatial locations combined with altered local field potentials provides a potential mechanism to explain spatial memory impairment after PAE.

Disclosures: R.E. Harvey: None. **J.K. Goss:** None. **T. Rigg:** None. **L.E. Berkowitz:** None. **L.M. Sanchez:** None. **J.L. Wagner:** None. **D.D. Savage:** None. **D.A. Hamilton:** None. **B.J. Clark:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.12/TT65

Topic: H.01. Animal Cognition and Behavior

Support: NIGMS (P30GM103400)

Title: Sex specific spatial navigation and spatial memory impairment in the TgF344-ad rat model of Alzheimer's disease

Authors: *L. E. BERKOWITZ, S. M. THOMPSON, E. N. DRAKE, J. T. MADDEN, E. A. SNEDDON, R. E. HARVEY, B. J. CLARK
Psychology, Univ. of New Mexico, Albuquerque, NM

Abstract: Spatial navigation and memory are impaired in early stages of Alzheimer's disease (AD), and may be a defining behavioral marker of preclinical AD. The advent of transgenic murine and rat models have proved valuable for aspects of neuropathological mechanisms in brain regions involved spatial cognition. Nevertheless, the translational significance resultant from these models may not represent that found in humans given limited pathological expression. However, the TgF344-AD rat model of AD expresses the full spectrum of AD pathology and may serve as a better rodent model for elucidating behavioral markers. In addition, TgF344-AD rats have been shown to exhibit reference memory impairment, though this has not been comprehensively assessed at early age points. This study aimed to characterize spatial memory in TgF344-AD at early ages and over time. TgF344-AD (n=16) and Fischer 344 (n=12) male and female rats were assessed in three paradigms of the Morris Water Maze at 4.3-5.5, 8.7-9.9 and 10.3-11.5 months of age. Measures of escape latency, preference score and search error were obtained. Navigational strategies characterized from swim paths were also obtained. Results indicate that TgF344-AD females exhibited navigational deficits as early as 4.5 months while TgF344-AD males showed impairments at 10.5 months. Furthermore, TgF344-AD males demonstrated acute reference memory impairments at 10.5 months whereas TgF344-AD females were no different from controls. Across all time points, cued navigation, spatial working memory and reference memory remained largely intact between subjects. Overall, these results indicated that TgF344-AD rats exhibit comparable deficits to those found in individuals with MCI and provides further evidence of sexual dimorphisms of AD. Future studies elucidating neurobiological mechanisms of spatial navigation impairment in preclinical AD would therefore benefit from using this model.

Disclosures: L.E. Berkowitz: None. **S.M. Thompson:** None. **E.N. Drake:** None. **J.T. Madden:** None. **E.A. Sneddon:** None. **R.E. Harvey:** None. **B.J. Clark:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.13/TT66

Topic: H.01. Animal Cognition and Behavior

Title: Brainwide coherent theta oscillations during visual and olfactory guided spatial learning and memory

Authors: *A. SHERIFF[1,2], L. M. KAY[1,2]
[1]Dept. of Psychology, [2]Inst. for Mind and Biol., The Univ. of Chicago, Chicago, IL

Abstract: Spatial learning and memory (SLM) has been widely studied in basic research of the hippocampus (HPC) primarily using visual cues. However, in navigation nocturnal rodents use multiple orientation mechanisms from multiple sensory sources, including odors. A recent proposal suggests that olfaction evolved primarily in the context of spatial navigation. Therefore, the rodent SLM model may be more ethologically valid when adding olfactory-spatial cues rather than using solely visual-spatial cues. Coherent neural oscillations underlie several component processes involved in SLM (e.g., perception, attention, and memory). However, while theta oscillations (4-14 Hz) are widely implicated in SLM, research on coherent oscillatory activity across widely distributed brain regions implicated in SLM is sparse. In this study, we used behavioral and electrophysiological methods with aims of 1: comparing SLM in rodents across visual and olfactory modalities; and 2: assessing the roles of coherent theta oscillations across olfactory-limbic and neocortical brain areas during SLM with olfactory vs. visual cues. We appetitively conditioned Long Evans rats to a single location within a four-arm radial maze to a learning criterion of 9 out of 10 correct trials using allocentric visual or olfactory cues. Preliminary data suggest rats learn the task faster when olfactory cues are present. Recordings of the local field potential (LFP) were made in olfactory-limbic and neocortical areas simultaneously during the navigation task: olfactory-limbic - olfactory bulb, piriform cortex, entorhinal cortex, HPC; neocortical - primary visual cortex, secondary motor cortex, medial parietal association cortex, primary somatosensory cortex. We performed within-subject comparisons of theta power and coherence during SLM. For electrophysiological analysis, rats were trained to a novel location each day for 4 consecutive days- two consecutive days with visual spatial cues and two consecutive days with olfactory spatial cues, with order counter-balanced across subjects. Theta coherence and power were analyzed in relation to SLM and correct vs. incorrect trials. Theta power increased in all regions while rats ran during the task, and V1M showed increased theta power during sessions with visual spatial cues. Theta coherence between pairs of brain regions fluctuated with SLM and in regards to correct vs. incorrect trials. Granger causality was used to assess directionality of coherence between all

pairs. Ubiquitous and modality-specific physiological features of SLM are elucidated by expanding analysis of SLM to olfactory cues and widely distributed theta coherence.

Disclosures: A. Sheriff: None. **L.M. Kay:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.14/UU1

Topic: H.01. Animal Cognition and Behavior

Support: NSERC CREATE

Natural Sciences and Engineering Research Council of Canada (NSERC) Discovery Grant #40352 (MHM)

Title: Spatiotemporal patterns of cortical voltage activity during sharp-wave ripples: Implications for memory consolidation theory

Authors: *J. KARIMI, M. NAZARI, B. L. MCNAUGHTON, M. H. MOHAJERANI
Neurosci., Canadian Ctr. For Behavioral Neuroscience/Univer, Lethbridge, AB, Canada

Abstract: The coordination of cortical up-states and hippocampal sharp-wave ripples (SWR) during slow wave sleep is believed to play a major role in the consolidation of recently acquired memories. Electrophysiological studies have extensively investigated the interaction of cortex and hippocampus during SWR, but the spatial distribution of SWR-up-state interaction across the cortical mantle is not known well. fMRI studies in monkeys have identified regions in the so-called default mode network which show high co-activity with SWRs (Kaplan et al., 2016), but fMRI suffers from the low temporal resolution. Therefore, we investigated how a large portion of the cortex interacts statistically with SWR using wide-field optical imaging of cortical voltage responses combined with extracellular recording of SWR and multi-unit activity (MUA) in CA1. Voltage-sensitive dye recording allowed us to capture cortical activity with high spatiotemporal resolution. Mice were imaged under urethane anesthesia as a model of slow wave sleep. We found that the activities of areas adjacent to the midline sinus, especially retrosplenial cortex (RSC) show the strongest correlation with SWRs. We also parsed the hippocampal activity around SWRs into 3 different clusters based on the pattern of hippocampal MUA activity and found distinct cortical response patterns corresponding to each of these groups. In general, the cortical activation tended to either lead, lag, or be coincident with the peaks of the SWR in these three clusters. Previously it was found that midline cortical reactivation of recent memory traces is strongest during periods of high up-down state transitions (Johnson et al., 2010). We speculate that the variable temporal relationships observed here may reflect the degree of consolidation of

different memory patterns: for well consolidated memory cortical activation may lead hippocampus whereas for the newest memories, that still depend on hippocampus, cortex would follow. This predicts a range of temporal interactions on average, which is what we observe.

Disclosures: J. Karimi: None. **M. Nazari:** None. **B.L. McNaughton:** None. **M.H. Mohajerani:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.15/UU2

Topic: H.01. Animal Cognition and Behavior

Support: FAPESP 2013/20378-8

Title: Differential genes expression following EGb treatment and conditioned suppression in the amygdaloid complex and dorsal hippocampal formation

Authors: *S. M. CERUTTI[1], C. R. ZAMBERLAM[2], J. M. CERUTTI[3]
[1]Univ. Federal De Sao Paulo, Sao Paulo, Brazil; [2]Federal Univ. of Sao Paulo, Diadema, Brazil; [3]Federal Univ. of Sao Paulo, São Paulo, Brazil

Abstract: Studies in our laboratory have shown that fear conditioning is associated with changes in gene expression of NMDA, 5-HT_{1A} and $GABA_A$ receptors in the dorsal hippocampal formation (dHF). This study assessed the effects of treatment prior to conditioning with EGb on differential *Grin2b,* Girn2a*5htr1a*, *Gfap* and *Gabraa*-α5 gene expression in the dorsal hippocampal formation (dHF) and amygdaloid complex (AC) of the rats submitted to the acquisition of the conditioned suppression of a lick response. Adult male *Wistar* rats were treated before conditioned fear with EGb or vehicle and were fear conditioned in a lick-operandum chamber. Each rat was placed in the chamber to acquisition of licking responses (1° to 5° days). In the 6°day, thirty minutes after the administration of drug or vehicle, each rat was submitted to four tone-footshock (CS-US) pairings. The suppression of licking response was calculated for each rat, in 10 trials CS presentation in the Acquisition Test (8°day). All rats had acquisition of fear memory but, in minor dose, EGb-treatment reduced the suppression ratio. The gene expression analysis, using qPCR, in the DH samples showed that 500 $mg.Kg^{-1}$ EGb treatment resulted in the overexpression of *Gabra5* ($Re_{Gabraa\text{-}\alpha5}$=268 ddCT), *Gabra1* ($Re_{Gabraa\text{-}\alpha1}$=4.879 ddCT), *Grin2b* ($Re_{Grin2b}$=382 ddCT) and *Gfap (*$Re_{Gfap}$=2.404 ddCT) in the DH after retention test when compared with control group (Re=676; 56; 180; 1268 and 63 respectly) (p<0,0001). In the AC, 500$mg.Kg^{-1}$ EGb resulted in upregulation of *5htr1a* (Re=187, p< 0.05) and *Gabraa*-α1 (Re=76; p< 0.0001) compared with control group (Re= 48 and 46, respectively). Conversely, 250 $mg.Kg^{-1}$ EGb treatment resulted in the downregulation of Re_{Grin2b}=0,1; $Re_{Gabraa\text{-}\alpha1}$=0,5 and

$Re_{Gabraa\text{-}\alpha5}$=1,7 ($p$<0,0001) in the AC when compared to the control group (Re=1,00; 4,6 and 13, respectively). Moreover, treatments with 1000 mg.Kg^{-1} EGb resulted in downregulation of $Re_{Gabraa\text{-}\alpha1}$=2,25 and $Re_{Gabraa\text{-}\alpha5}$=7,3; p<0,0001). One-way ANOVA followed by a *post hoc* Bonferroni test was performed to evaluate the relationships between the expression levels and groups. The results from gene expression analysis in the DH and AC shown the modulatory effects of EGb treatment and indicate the involvement of 5HT1AR, GABAA and NMDA-GluN2B receptors in the effect of standardized extract of *Ginkgo biloba* L. on acquisition of conditioned fear in a dose-dependent and structure-dependent manner (CEUA N. 3298260514).

Disclosures: S.M. Cerutti: None. **C.R. Zamberlam:** None. **J.M. Cerutti:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.16/UU3

Topic: H.01. Animal Cognition and Behavior

Support: NSF Career Award 1565410 (IAM)

NIH Grant 1SC1GM122645 (AjA)

Title: Physiological characteristics and functional role of ventral hippocampus projecting cells

Authors: *M. R. LOPEZ[1], H. ZURITA[1], B. HARLAND[2], K.-C. LEONG[1], A. J. APICELLA[1], I. A. MUZZIO[1]
[1]Biol., Univ. of Texas At San Antonio, San Antonio, TX; [2]Psychology, Univ. of Arizona, Tucson, AZ

Abstract: Many psychopathologies such as post-traumatic stress disorder and specific phobias are typified by failures of extinction and fear generalization. Therefore, understanding the underling circuit abnormalities that lead to these psychopathologies is of critical importance. The hippocampus is thought to provide a contextual “gating” mechanism that determines whether fear or extinction should be expressed in particular contexts; however, no direct physiological evidence supports this theory. The ventral hippocampus (VH) is in a key position to control this contextual “gating” through its projections to the basolateral amygdala (BLA), an area involved in associative fear learning, and the prelimbic (PL) and infralimbic (IL) cortices, regions in the medial prefrontal cortex (mPFC) involved in fear expression and extinction, respectively. However, since single cells in the VH have broadly tuned fields (e.g., cells fire in large areas of space), it is unclear how they provide specific contextual information during learning and retrieval. Our lab recently demonstrated that although single VH cells lack precise spatial tuning, the population activity in the VH contains contextual information. Critically, ventral patterns of

activity are ideal to support generalization processes. This raises the question of whether the VH projection neurons are comprised by distinct cell-types and/or provide the same computational information to the different target regions of the fear/extinction circuitry. To test this, we first used retrograde fluorescent beads to identify ventral projecting cells to mPFC and BLA. We found that these different projecting cells conform segregated cell-types with almost no overlap. Then, we began to characterize their physiological and anatomical properties using whole cell patch clamp. Our preliminary data suggest that ventral cells projecting to mPFC burst and display temporal summation, whereas cells projecting to amygdala display regular firing and no temporal summation. We are currently trying to elucidate the distribution and electrophysiological properties of cells projecting to PL and IL cortices. To further characterize the functional role of VH in generalization processes, we are also using chemogenetic silencing of specific projecting cells in combination with single unit recordings from ventral area CA1. These results will provide important information about how the VH provides contextual gating of fear- and extinction- related behaviors.

Disclosures: **M.R. Lopez:** None. **H. Zurita:** None. **B. Harland:** None. **K. Leong:** None. **A.J. Apicella:** None. **I.A. Muzzio:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.17/UU4

Topic: H.01. Animal Cognition and Behavior

Support: NIH Early Independence Award (DP5)

NARSAD

Ludwig Family Foundation

Milton Fund

Title: Chronically reactivating positive and negative memories to modulate hedonic and social behaviors

Authors: *E. DOUCETTE, E. MERFELD, Y. ZAKI, S. L. GRELLA, N. J. MURAWSKI, M. SHPOKAYTE, S. RAMIREZ
Ctr. for Brain Sci., Harvard Univ., Cambridge, MA

Abstract: Chronic stress in mice induces variety a of anxiety- and depressive-like phenotypes at the neuronal, circuit, and behavioral levels. More specifically, social and hedonic-like states are dramatically impaired across many psychiatric disorders, though the underlying mechanisms

sufficient to precipitate or alleviate such impairments remain largely unknown. Here, we utilize an activity-dependent and inducible tagging strategy to modulate hippocampus cells processing positive, neutral, or negative memories prior to chronic immobilization stress exposure. We hypothesized that chronic positive memory stimulation is sufficient to mitigate the effects of stress, while chronic negative memory stimulation is sufficient to mimic such effects in a number of behavioral assays (e.g. exposure to female, resident intruder, social interaction). We find that chronically stimulating hippocampus cells processing a positive memory in male mice is sufficient to increase hedonic-like activity, as measured by time spent interacting with a female mouse, and those effects remain unaltered by chronic stress. We also find that chronic stimulation of cells processing a negative memory is sufficient to impair social behaviors in both the resident intruder and social interaction assays. The magnitude of these effects is comparable to the impairments seen in chronically stressed animals receiving neutral memory stimulation pre-stress. Furthermore, chronically stimulating cells processing a negative memory followed by chronic stress produces larger impairments than stress alone in a subset of social behaviors. Our current experiments focus on interrogating the neural circuitry underlying each phenotype by measuring levels of neurogenesis, dendritic spine growth and retraction, and circuit-wide activity contributing to hedonic and social behaviors. Together our results connect chronic memory modulation with discrete maladaptive behavioral states and simultaneously provide a mechanistic basis for inducing a defined set of behavioral impairments.

Disclosures: **E. Doucette:** None. **E. Merfeld:** None. **Y. Zaki:** None. **S.L. Grella:** None. **N.J. Murawski:** None. **M. Shpokayte:** None. **S. Ramirez:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.18/UU5

Topic: H.01. Animal Cognition and Behavior

Support: NIH Early Independence Award (DP5)

NARSAD

Ludwig Family Foundation

Milton Fund

Title: Inhibiting ensembles in the hippocampus and amygdala to suppress reinstatement-induced fear

Authors: *Y. ZAKI, E. DOUCETTE, S. L. GRELLA, N. J. MURAWSKI, E. MERFELD, M. SHPOKAYTE, S. RAMIREZ
Ctr. for Brain Sci., Harvard Univ., Cambridge, MA

Abstract: Post-traumatic stress disorder (PTSD) is a condition that precipitates from a highly aversive experience and is manifested by overgeneralized fear in innocuous situations. Interestingly, a striking proportion of patients who undergo exposure therapy - which can lead to the suppression, or "extinction," of the original fear memory - are highly vulnerable to relapse, especially when the conditioned stimulus is delivered outside a clinical context. Here, we seek to interrogate the neural substrates supporting the acquisition of fear as well as the subsequent extinction of fear to gain a causal understanding of its underlying neural components. We used a combination of activity-dependent labeling of neuronal ensembles in multiple brain regions associated with fear-related behaviors (basolateral amygdala, BLA; dorsal dentate gyrus of hippocampus, dDG) and further manipulated these ensembles using optogenetics to probe the changes that a fear memory undergoes during extinction and during fear reinstatement.
We first tagged BLA cells processing a contextual fear memory in mice. All subjects then underwent extinction learning, and had the tagged fear ensemble in the BLA inhibited either during a shock in an unconditioned context (serving to reinstate the original context-specific fear) or during a fear recall test in the original conditioned context the day after. We found that while inhibition of the original fear ensemble in the BLA was not enough to prevent reinstatement, inhibition of that ensemble during the recall test was enough to actively disrupt fear expression in the conditioned context. This suggests that the original fear ensemble in the BLA contributes to a context-dependent fear response following shock-induced reinstatement, providing key evidence that the neural correlate of fear reinstatement may be a reemergence of the original fear memory trace. In the dDG, we observed that the original fear ensemble was preferentially reactivated during the fear recall session, but not during reinstatement. To further investigate the changes that occur in the brain during reinstatement, our current work focuses on inhibiting the hippocampal fear ensemble during the fear recall test to assess whether that ensemble continues to be instrumental in the contextual fear response following reinstatement.

Disclosures: Y. Zaki: None. **E. Doucette:** None. **S.L. Grella:** None. **N.J. Murawski:** None. **E. Merfeld:** None. **M. Shpokayte:** None. **S. Ramirez:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.19/UU6

Topic: H.01. Animal Cognition and Behavior

Support: NIH Early Independence Award (DP5)

NARSAD

Ludwig Family Foundation

Milton Fund

Title: Artificially enhancing or suppressing hippocampus-mediated fear memories

Authors: *N. J. MURAWSKI[1], B. CHEN[2], S. L. GRELLA[1], M. SHPOKAYTE[1], E. MERFELD[1], Y. ZAKI[1], E. DOUCETTE[1], S. RAMIREZ[1]
[1]Ctr. for Brain Sci., Harvard Univ., Cambridge, MA; [2]Neurobio. and Behavior, Columbia Univ. Med. Ctr., New York, NY

Abstract: Cognitive and emotional disruptions underlie many stress disorders and are also known to affect hippocampus function. The dorsal hippocampus is thought to processes spatial, temporal, and contextual information whereas the ventral hippocampus is thought to mediate stress and emotional aspects of learning and memory. Here, we asked if artificial activation of positive or negative memories in the dorsal or ventral hippocampus could promote appetitive or aversive-related behaviors. We also examined if chronic activation of a fear memory is sufficient to mimic extinction-like reductions in fear responses. We first infused an activity-dependent AAV9-c-Fos-tTA-TRE-ChR2 into the dorsal or ventral dentate gyrus (DG), which allowed for optical control over DG neurons expressing the immediate-early gene c-Fos in a doxycycline(Dox)-dependent manner. Mice received a positive, negative, or neutral experience while off Dox to tag cells processing a discrete memory. Next, mice underwent a battery of behavioral tests to measure appetitive and aversive responses during light-on and light-off epochs. Our findings are threefold: 1) activation of dorsal and ventral hippocampus cells were sufficient to drive freezing, avoidance, and preference; 2) the ventral, but not dorsal, hippocampus was sufficient to modulate anxiety-like states; 3) and finally, chronic activation of the dorsal hippocampus cells produced extinction-like reductions in fear responses, whereas ventral hippocampus stimulation induced a context-specific enhancement of a fear memory. Together, we demonstrate functionally overlapping and segregated roles of the dorsal and ventral hippocampus and that artificially activating positive and negative memories are sufficient to mitigate or mimic stress-related behavioral states.

Disclosures: **N.J. Murawski:** A. Employment/Salary (full or part-time):; Harvard University. **B. Chen:** None. **S.L. Grella:** None. **M. Shpokayte:** None. **E. Merfeld:** None. **Y. Zaki:** None. **E. Doucette:** None. **S. Ramirez:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.20/UU7

Topic: H.01. Animal Cognition and Behavior

Support: NIH Early Independence Award (DP5)

NARSAD

Ludwig Family Foundation

Milton Fund

Title: Activating ventral hippocampus to amygdala terminals processing fear and reward

Authors: *M. SHPOKAYTE, S. L. GRELLA, Y. ZAKI, N. J. MURAWSKI, E. DOUCETTE, E. MERFELD, S. RAMIREZ
Ctr. for Brain Sci., Harvard Univ., Cambridge, MA

Abstract: Projections from the hippocampus to the amygdala code for a variety of contextual information associated with positive and negative valences. Previous studies have focused on stimulating cell bodies in the hippocampus or amygdala during events of opposing valences and have revealed that a discrete set of hippocampus cells can "switch" which valence-specific behavior they drive, while such responses in the amygdala appear to be more hardwired. However, the flexibility of hippocampus-amygdala terminals in processing both reward and aversion remains untested. Here, we utilize the activity dependent and inducible cfos-tTA system for tagging cells processing positive or negative memories in ventral CA1 terminals onto the basolateral amygdala of c57BL/6 mice. Following tagging, the capability of these terminals to drive preference or aversion was assessed. Next, the animals were exposed to either a negative or positive experience while the hippocampus terminals processing the opposite valence were optogenetically activated. We find that optogenetic stimulation of hippocampus to amygdala terminals is sufficient to switch the terminals' capacity to drive reward to fear and vice versa. Our ongoing experiments are focused on anatomically tracing projection-specific targets of positive and negative memory hippocampus terminals and testing their role in modulating a battery of behavioral states. Understanding the intricacies of the hippocampus to amygdala circuitry in relation to the malleability of memories may have clinical value for the treatment of psychiatric disease-related states.

Disclosures: M. Shpokayte: None. **S.L. Grella:** None. **Y. Zaki:** None. **N.J. Murawski:** None. **E. Doucette:** None. **E. Merfeld:** None. **S. Ramirez:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.21/UU8

Topic: H.01. Animal Cognition and Behavior

Support: NIH Early Independence Award (DP5)

NARSAD

Ludwig Family Foundation

Milton Fund

Title: Reactivating hippocampus-mediated memories to disrupt the reconsolidation of fear

Authors: *S. L. GRELLA, Y. ZAKI, N. J. MURAWSKI, E. DOUCETTE, E. MERFELD, M. SHPOKAYTE, S. RAMIREZ
Ctr. for Brain Sci., Harvard Univ., Cambridge, MA

Abstract: Background: Fear conditioning has been used to model memory and stress-related behaviors in rodents. While fear is often adaptive, dysregulation of fear circuits can lead to maladaptive states comprising mood and anxiety disorders. A promising prospect of attenuating the strength of fear memories is through the disruption of reconsolidation - a process by which activated memories are susceptible to modification, thus allowing for subsequent manipulation (e.g. enhancement / disruption). In previous studies, interventions such as protein synthesis inhibitors and beta-blockers have been used to target and disrupt conditioned fear during the reconsolidation process. Here, we propose a novel intervention based on the hypothesis that optogenetic reactivation of a previously formed, hippocampus-mediated memory during reconsolidation will alter and disrupt the original fear memory, thereby reducing the behavioral expression of fear. **Method:** We combined the use of the activity-dependent inducible c-Fos-tTA system for neuronal tagging in c57BL/6 mice to tag dorsal dentate gyrus (dDG) cells active during a positive or neutral experience, and channelrhodopsin 2-mediated optogenetics. Mice were fear-conditioned and given a 20-minute recall session the following day. During recall, we optically stimulated the dDG during the first or last 10 minutes of the session when the reconsolidation window is thought to be open – to reactivate the tagged dDG-mediated positive or neutral memories. Mice then received extinction training and were tested for stress-induced reinstatement on subsequent days. **Results:** Artificial reactivation of both positive and neutral memories during reconsolidation resulted in faster rates of extinction learning and the attenuation of stress-induced reinstatement. However, these effects transpired more quickly with the reactivation of a positive memory compared to the reactivation of a neutral memory. Moreover, artificial reactivation of a positive memory during reconsolidation of a fear memory resulted in real-time decreases in freezing during reactivation whereas reactivation of a neutral memory did not produce similar decreases in freezing. **Significance:** This work highlights the therapeutic value of memory modulation as a viable treatment for the suppression of fear responses, implicating dDG cells as specific nodes of intervention.

Disclosures: S.L. Grella: None. **Y. Zaki:** None. **N.J. Murawski:** None. **E. Doucette:** None. **E. Merfeld:** None. **M. Shpokayte:** None. **S. Ramirez:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.22/UU9

Topic: H.01. Animal Cognition and Behavior

Support: NIH Grant R01 MH100318

Title: Neuronal activity in the amygdala and hippocampus during and after optogenetic stimulation of the basolateral amygdala in awake rats

Authors: *N. S. AHLGRIM[1,2], C. R. GALLOWAY[1], Y. CHUNG[3], K. PARK[3], J. R. MANNS[1]
[1]Psychology, [2]Grad. Program in Neurosci., [3]Emory Univ., Atlanta, GA

Abstract: The amygdala is known to modulate memory consolidation via its direct and indirect projections to the hippocampus. Although this modulation is most often studied in conjunction with emotional arousal, like during fear conditioning and acute stress, recent work has shown that activity in the basolateral amygdala (BLA) can also modulate emotionally neutral memory. We have previously found that brief electrical stimulation of the BLA enhances recognition memory for individual objects, a phenomenon that is dependent on the intermediate hippocampus. Further, electrical stimulation to the BLA increases synchrony between the CA3 and CA1 subfields of the hippocampus. To better characterize the activity in the BLA that produces the enhancement of memory and to determine whether these effects are attributable to activation of glutamatergic projection neurons in the BLA, the current study used optogenetic stimulation in place of electrical stimulation. Optogenetic stimulation facilitated simultaneous recordings of single-unit and oscillatory activity in the amygdala and hippocampus. Putative glutamatergic neurons were transfected with the depolarizing Channelrhodopsin [ChR2(H134R)] ion channel, and transfected neurons were stimulated across a range of frequencies. Simultaneous stimulation and recording in the amygdala allows for on-line verification of how amygdala activity changed as a result of optical stimulation. Optogenetic stimulation of the BLA elicited spiking and oscillations in local field potentials that were entrained to the stimulation frequency. Our protocol of simultaneous neural recordings from the amygdala and hippocampus in an awake animal will enable direct analysis of single-unit and oscillatory activity in the amygdala during memory tasks, which projects to and modulates the hippocampus. The current experiment lays the foundation for the long-term goal of the project, which is to understand how these two regions communicate and what neural activity is required to enhance the consolidation of specific memories.

Disclosures: N.S. Ahlgrim: None. **C.R. Galloway:** None. **Y. Chung:** None. **K. Park:** None. **J.R. Manns:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.23/UU10

Topic: H.01. Animal Cognition and Behavior

Support: NIH 2R01MH080007

WaNPRC Base Grant P51 OD010425

Title: Conjunctive coding in the primate entorhinal cortex

Authors: *M. L. MEISTER[1,2], E. A. BUFFALO[1,2]
[1]Physiol. and Biophysics, [2]Natl. Primate Res. Ctr., Univ. of Washington, Seattle, WA

Abstract: Primate entorhinal cortex neurons respond to the appearance of visual stimuli in the context of visual recognition memory or association tasks in which the neural responses are typically analyzed during time periods lasting about 0.5 seconds. Given the heterogeneity of entorhinal response types identified *in vitro,* including responses that continue many seconds after stimulation, it is possible that distinct cell classes could also be observed *in vivo* across longer time periods. Here, we recorded the responses of individual neurons (n = 349) while monkeys freely viewed large, natural scene stimuli. Monkeys were allowed to view the scenes for up to 5 seconds, and up to 120 distinct scenes were presented within each session. Results revealed that entorhinal neurons exhibit vastly distinct responses locked to the onset of visual stimuli, both enhanced and reduced firing rates, with some neurons returning to baseline firing rate quickly, and others showing an exceptionally long decay, continuing well over 1 second after stimulus onset. Responses spanning this dynamic range were reliably encountered across experimental sessions and animals, suggesting distinct cell types.
To determine whether these separate cell types reflected anatomical specificity or differed in other aspects of their response profile, we compared the properties of cells with “Brief” vs. “Sustained” responses (responses that respectively returned to baseline firing rate within 0.5 sec or no sooner than 1.5 sec following image onset). Compared to Brief cells, Sustained cells included a significantly greater proportion of cells located in superficial cortical layers which provide the predominant cortical input to the hippocampus. Additionally, Sustained cells were more likely to show consistent spatial activity that reflected the location of the monkey’s gaze within the image. Notably, there was no difference in the proportion of spatial Sustained cells that showed enhanced compared to reduced firing rate at stimulus onset.
Together, these findings reveal an association between cells with an exceptionally long response decay after the appearance of an image, spatial coding, and location in the superficial cortical layers. Because the entorhinal cortex is important for memory, we speculate that these exceptionally long responses may provide necessary drive to create a sustained binding state, or

memory “chunk,” linking across time spatially separate elements encountered during visual exploration of an image. Future research distinguishing these diverse cell types offers a potentially powerful, new way to enhance our understanding of the neural mechanisms that underlie memory.

Disclosures: M.L. Meister: None. **E.A. Buffalo:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.24/UU11

Topic: H.01. Animal Cognition and Behavior

Support: NIH Grant 2R01MH080007

NIH Grant R01MH093807

WaNPRC Base Grant P51 OD010425

Pfizer, Inc.

Title: Mnemonic discrimination task in rhesus macaques

Authors: *C. I. O'LEARY[1,2], M. L. JUTRAS[1,2], A. NG[1,2], S. A. SCHLEUFER[1,2], A. J. O. DEDE[1], Z. REAGH[3,4], M. A. YASSA[3,4], E. P. LEBOIS[5], E. A. BUFFALO[1,2]
[1]Physiol. and Biophysics, [2]Washington Natl. Primate Res. Ctr., Univ. of Washington, Seattle, WA; [3]Ctr. for the Neurobio. of Learning and Memory, Univ. of California Irvine, Irvine, CA; [4]Neurobio. and Behavior, Univ. of California, Irvine, Irvine, CA; [5]Intrnl. Med. Res. Unit, Pfizer, Inc., Cambridge, MA

Abstract: Pattern separation, which refers to the process of orthogonalizing overlapping inputs into distinct memory representations, has been shown to be affected in healthy elderly individuals as well as in individuals with mild cognitive impairment who may go on to develop Alzheimer’s disease. Research using functional imaging in humans has implicated substructures of the hippocampus, specifically CA3 and Dentate Gyrus (DG), as being particularly important for pattern separation, with deficits in nondemented older adults associated with increased CA3/DG activity (Lacy, et al., 2011; Yassa et al., 2010). Single unit recordings in rodents have implicated the DG in pattern separation and have suggested that CA3 representations more likely reflect pattern completion mechanisms, i.e., the retrieval of a memory with partial input (Neunuebel and Knierim, 2014). Monkeys, which can be trained to perform similar, if not identical, behavioral tasks as those used in human subjects, provide an avenue for clarifying this issue and assessing the neurobiological underpinnings of this ability at the level of cells and

circuits. Here, we adapted the mnemonic discrimination task from the human literature for use with rhesus macaques. This task provides a useful behavioral assay of pattern separation. Five monkeys were trained on a sequential delayed match to sample task with centrally located images. Subjects indicated the detection of a match by touchbar release. Each trial could include repeated nonmatching stimuli (ABBA design) and extend to a potential length of six nonmatching stimuli. Human-validated distracter items from the human mnemonic discrimination task that varied parametrically across five levels of similarity to the target item were used as critical lure stimuli. All monkeys were able to perform this task successfully. Notably, monkeys displayed similar behavioral performance to humans, in that performance varied predictably along with the increasing similarity of the lure to the target. These results demonstrate the applicability of the mnemonic discrimination task as a platform for further neurophysiological studies targeting single units in specific hippocampal subregions in the primate.

Disclosures: **C.I. O'Leary:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Partial support provided by Pfizer, Inc. **M.L. Jutras:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Partial support provided by Pfizer, Inc. **A. Ng:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Partial support provided by Pfizer, Inc. **S.A. Schleufer:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Partial support provided by Pfizer, Inc. **A.J.O. Dede:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Partial support provided by Pfizer, Inc.. **Z. Reagh:** None. **M.A. Yassa:** None. **E.P. Lebois:** A. Employment/Salary (full or part-time):; Employee of Pfizer, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); I own Pfizer, Inc. Stock. **E.A. Buffalo:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Partial support provided by Pfizer, Inc..

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.25/UU12

Topic: H.01. Animal Cognition and Behavior

Support: NIH R01MH093807

WaNPRC Base Grant P51 OD010425

NIH DA032466

NIH DA032436

Title: Eye movements temporally organize spatial representations in the primate hippocampus

Authors: *S. D. KOENIG[1], E. A. BUFFALO[2,3]
[2]Physiol. and Biophysics, [3]Washington Natl. Primate Res. Ctr., [1]Univ. of Washington, Seattle, WA

Abstract: Single unit recordings in rodents have identified place and time cells in the hippocampus that are hypothesized to support the spatial and temporal aspects of episodic memories. However, it is not fully understood the extent to which similar representations exist in the primate hippocampus. Notably, studies in monkeys have identified neurons in the hippocampus and entorhinal cortex that are modulated by gaze location rather than the monkey's physical location (Rolls, 1999; Killian et al., 2012). In addition, saccadic eye movements modulate the activity of single hippocampal neurons (Sobotka et al., 1997) and the phase of the hippocampal local field potential (Hoffman et al., 2013; Jutras et al., 2013), suggesting that the temporal properties of active sampling of the environment via saccades plays an important role in hippocampal activity. To further explore how these representations are related to eye movements, we recorded the activity of 347 hippocampal neurons from two monkeys as they freely viewed complex images. We found that 31% of these neurons were significantly modulated by viewing location; these neurons were spatially stable across the recording session and had significant ($p < 0.05$) spatial information scores.(Skaggs et al., 1993) Surprisingly, each view-modulated neuron appeared to reach its maximum firing rate at a distinct time relative to the start of a fixation into the neuron's firing field. Across the population of view-modulated neurons, the latency of the peak response tiled the full duration of a fixation (~200 msec), suggesting that these responses may reflect sequential firing. Taken together, these data provide further support for the idea that neurons in the primate hippocampus are modulated by eye movements and show selectivity for viewing location. Furthermore, these data demonstrate that

eye movements temporally organize hippocampal activity across the extent of a fixation, potentially allowing information to be integrated across successive eye movements.

Disclosures: S.D. Koenig: None. **E.A. Buffalo:** None.

Poster

425. Learning and Memory: Hippocampal-Parahippocampal-Limbic Interactions

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 425.26/UU13

Topic: H.01. Animal Cognition and Behavior

Support: NIH Training Grant in Computation Neuroscience 5T90DA032436

NIH 2R01MH080007

Simons Foundation 325467

Simons Foundation 368149

WaNPRC Base Grant P51 OD010425

Washington Research Foundation Innovation Graduate Fellowship in Neuroengineering

Washington Research Foundation University of Washington Institute for Neuroengineering (UWIN)

Title: Context-specific representations in monkey hippocampal neurons

Authors: *Y. BROWNING[1], J. W. RUECKEMANN[2], K. L. MORRISROE[3], S. A. SCHLEUFER[2], M. J. JUTRAS[2], A. L. FAIRHALL[2,4], E. A. BUFFALO[2,3]
[1]Grad. Program in Neurosci., [2]Physiol. and Biophysics, [3]Washington Natl. Primate Res. Ctr., [4]Inst. for Neuroengineering, Univ. of Washington, Seattle, WA

Abstract: Decades of studies have shown that the hippocampus plays an important role in memory in humans, monkeys, and rodents, but we do not yet have a full understanding of how transient neuronal activity in the hippocampus supports mnemonic function. Here, we investigated activity in the monkey hippocampus related to spatial memory performance. Monkeys were trained on a spatial delayed-alternation task in a visually rich virtual Y-Maze, in which the previous trajectory must be remembered in order to make a correct response. Hippocampal neurons were simultaneously recorded from more than 60 channels using a chronically implanted microdrive with independently movable electrodes (Gray Matter Research). The electrodes targeted the full anterior-posterior and medial-lateral extent of the

hippocampus, spanning the dentate gyrus, CA3, CA1, and subliculum.
Preliminary results indicate that single units show reliable responses to multiple distinct features of the task. Hippocampal neurons demonstrate discrete spiking in each phase of the task, including the center stem, choice point, reward locations, and delay period. Notably, these neural representations include place cells with consistent responses to the position of the monkey's virtual avatar relative to maze landmarks. Over the aggregate ensemble, the observed hippocampal activity spans all task events, bridging discrete features within one network.
To determine whether visual features of the maze influence network activity in addition to task structure, we compared neuronal activity across distinct environments with the same spatial dimensions. Neural responses were strikingly consistent across repeated sessions in a particular environment during a single day. When a visually distinct environment was interposed between the two repeated environments, a subpopulation of hippocampal cells showed marked shifts in response profile. These alterations included changes in whether a neuron was active in the task and in the location of a neuron's place field. Taken together, these findings show that robust, task dependent activity in the monkey hippocampus can be elicited using a virtual reality memory paradigm. Further, environmental context biases which neural ensemble will be active, even with a consistent task structure.

Disclosures: Y. Browning: None. **J.W. Rueckemann:** None. **K.L. Morrisroe:** None. **S.A. Schleufer:** None. **M.J. Jutras:** None. **A.L. Fairhall:** None. **E.A. Buffalo:** None.

Poster

426. Learning and Memory: Hippocampal CA2 and Social Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 426.01/UU14

Topic: H.01. Animal Cognition and Behavior

Support: NSF DGE-16-44869

Title: Modified firing in hippocampal area CA2 in a mouse model of schizophrenia during social tasks

Authors: *M. L. DONEGAN[1], F. STEFANINI[1], Y. ZAFRINA[2], S. FUSI[1], J. A. GORDON[3], S. A. SIEGELBAUM[1]
[1]Neurosci., Columbia Univ., New York, NY; [2]Barnard Col., New York, NY; [3]Office of the Director, Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: Social interaction and bonding are crucial elements of mammalian survival, providing a support system for offspring survival, offering increased security, and reducing the expenditure of resources. Neuropsychiatric diseases, such as autism and schizophrenia, often present with disruptions in social behavior that are poorly understood. Recent work on area CA2 of the

hippocampus has shown that CA2 is necessary for the formation of social memory. Moreover, brains from individuals with schizophrenia show specific reductions in parvalbumin-positive (PV) interneurons in CA2. Despite these findings linking CA2 to social memory, the neural firing patterns underlying these memories have yet to be understood. The Df(16)A+/-mouse line is a mouse model of schizophrenia that recapitulates a microdeletion on the 22q11 chromosome, which confers a 30-fold increased likelihood of developing schizophrenia in humans. These mice show deficits in social memory, as well as an age-dependent loss of PV interneurons specific to CA2 (similar to the human results), and a hyperpolarization of the CA2 pyramidal neuron resting potential recorded in slices. While these physiological changes have been investigated in vitro, their in vivo consequences on CA2 firing have not been examined.To characterize CA2 firing during social behavior we recorded extracellularly from the CA2 region of both wild-type and Df(16)A+/- mice during a modified 3 chamber social interaction task during which the mice interact with 1) an empty chamber 2) novel objects 3) littermates and 4) novel mice. In wild-type animals, CA2 firing is highly heterogeneous, with CA2 spatial firing patterns highly unstable from session to session. Interestingly, about 10% of cells in CA2 are only active during the social sessions. A different subpopulation shows is an overall firing rate increase around the novel animal. Despite altered inhibition and resting potential, CA2 pyramidal cells from Df(16)A+/- mice show no change in either average or peak firing rate during this task. Interestingly, CA2 spatial firing patterns of the Df(16)A+/- mice are more stable across the different sessions, with the cells having fewer firing fields compared to their wild-type counterparts. Such changes in single cell firing patterns may interfere with the ability to decode novelty, social information, and spatial information from CA2 population activities and therefore ultimately to extract this information from downstream neurons. These findings suggest a possible role for CA2 neuronal activity in social memory, and that changes in CA2 firing may play a role in the social deficits seen in the Df(16)A+/- mice.

Disclosures: M.L. Donegan: None. **F. Stefanini:** None. **Y. Zafrina:** None. **S. Fusi:** None. **J.A. Gordon:** None. **S.A. Siegelbaum:** None.

Poster

426. Learning and Memory: Hippocampal CA2 and Social Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 426.02/UU15

Topic: H.01. Animal Cognition and Behavior

Support: 5 T32 MH 15174 39

R01MH104602-04

Title: Organization of CA2 projections to ventral CA1 neurons

Authors: *E. W. BUSS, F. LEROY, S. A. SIEGELBAUM
Dept. of Neurosci., Columbia Univ., New York, NY

Abstract: Axons of CA2 pyramidal neurons have a diverse pattern of innervation, providing synaptic input both onto dorsal CA1 (dCA1) as well as ventral CA1 (vCA1) neurons. Additionally, dorsal Ca2 (dCA2) and vCA1 pyramidal neurons are important for social memory processing and storage; however, the functional synaptic connectivity of this circuit remains poorly understood. Given the evidence of distinct functional domains along the dorsoventral axis in CA1 as well as cell type heterogeneity among CA1 pyramidal neurons, we sought to determine how these projections are integrated into the circuit. Using optogenetic stimulation of CA2 fibers, we characterized strong, excitatory projections to the vCA1, displaying a distinct pattern of innervation. Although it remains unclear how CA2 output influences behavior, this offers a potential social memory circuit.

Disclosures: E.W. Buss: None. **F. Leroy:** None. **S.A. Siegelbaum:** None.

Poster

426. Learning and Memory: Hippocampal CA2 and Social Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 426.03/UU16

Topic: H.01. Animal Cognition and Behavior

Support: NIH Grant R01 MH104602-01

Title: CA2 circuits controlling social behaviors

Authors: *F. LEROY[1], J. PARK[1], T. MEIRA[1,2], E. W. BUSS[1], S. A. SIEGELBAUM[1]
[1]Neurosci., Columbia Univ., New York, NY; [2]Life and Hlth. Sci. Res. Inst. (ICVS),, Sch. of Medicine, Univ. of Minho, Braga, Portugal

Abstract: The hippocampal CA2 area lodged between CA3 and CA1 integrates inputs from the entorhinal cortex, the dentate gyrus, CA3 as well as the supramammillary nucleus, the medial septum and the raphe. Its output remains, however, poorly understood. We characterized strong CA2 projections to the ventral CA1 area (vCA1) and the lateral septum (LS) and have been probing their respective behavioral function by silencing each set of terminals. CA2 and vCA1 were recently shown to be necessary for social memory. We find that dorsal CA2 projects to and excites ventral CA1, providing a potential social memory circuit. We further find that dorsal CA2 excites dorsal LS (dLS). Moreover this projection is not involved in social memory but acts to enhance social aggression through a disinhibitory circuit: CA2 excites dLS, which inhibits ventral lateral septum (vLS), thereby relieving tonic inhibition of the aggression-promoting hypothalamic VMHvl area. This is agreement with studies showing that deletion of the arginine

vasopressin 1b receptor (AVPR1b), which is highly enriched in CA2, reduces social aggression. Our results show that activation of AVPR1b on CA2 presynaptic terminals enhances CA2-LS excitatory synaptic transmission and that selective blockade of these receptors inhibits social aggression. Although it remains unclear how CA2 outputs and their modulations by vasopressin control different social behaviors, our study, together with previous results, suggests a close relationship between social memory and aggression. We are currently investigating the behavioral effect of silencing the CA2-vCA1 projection to explore whether differential neuromodulation of divergent projections from a single brain region may selectively recruit memory versus aggressive social behaviors.

Disclosures: F. Leroy: None. **J. Park:** None. **T. Meira:** None. **E.W. Buss:** None. **S.A. Siegelbaum:** None.

Poster

426. Learning and Memory: Hippocampal CA2 and Social Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 426.04/UU17

Topic: H.01. Animal Cognition and Behavior

Support: NIH Grant 5R01MH104602-04

Portuguese Foundation for Science and Technology MD/PhD scholarship PD/BD/113700/2015

Title: The hippocampal CA2 region plays a dynamic role in social memory

Authors: *T. MEIRA[1,3], F. LEROY[1], S. SIEGELBAUM[1,2]
[1]Neurosci., [2]Pharmacol., Columbia Univ., New York, NY; [3]Life and Hlth. Sci. Res. Inst. (ICVS), Sch. of Medicine, Univ. of Minho, Braga, Portugal

Abstract: The hippocampus is an extensively studied structure with a well-defined role in declarative memory. Studies over the past decade have revealed specific roles of certain hippocampal subregions in different aspects of memory storage and recall. Nevertheless, the function of the CA2 field is largely unknown. Recently, the importance of CA2 in social memory was directly demonstrated using a genetically engineered mouse line that allowed for the selective silencing of CA2 pyramidal neurons (PyNs). However, this approach depended on the irreversible and long-term inactivation of CA2 through expression of the tetanus toxin light chain, which may have led to long-term effects on neural circuitry outside of CA2 that consequently caused the social cognition deficits. To further explore how CA2 is acutely involved in social memory, we have been using pharmacogenetic tools that enable us to control the activity of CA2 in a defined temporal manner. We expressed the inhibitory hM4Di

DREADD selectively in CA2 PyNs by injection of a Cre-dependent AAV into the CA2 region of adult *Amigo2*-Cre mice, where Cre expression is largely restricted to CA2 PyNs. In acute hippocampal slices from viral-injected mice, application of the DREADD agonist clozapine *N*-oxide (CNO) reliably hyperpolarized the PyNs membrane, reducing excitability. We first examined the effects of acute CA2 silencing using the direct interaction test of social memory, in which a subject mouse is exposed to a novel juvenile stimulus mouse for 2 min in trial 1 and then re-exposed to the same stimulus mouse in trial 2 after a 30 min inter-trial interval. Social memory is normally manifest as a decrease in social exploration from trial 1 to trial 2 as a result of decreased social novelty. Intraperitoneal injection of CNO 30 min prior to the test caused a profound decrease in social memory, demonstrating the importance of dynamic on-line CA2 activity. Experiments are currently underway to examine whether CA2 is selectively required for memory storage, consolidation and/or retrieval.

Disclosures: T. Meira: None. **F. Leroy:** None. **S. Siegelbaum:** None.

Poster

426. Learning and Memory: Hippocampal CA2 and Social Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 426.05/UU18

Topic: H.01. Animal Cognition and Behavior

Support: Skirball Insitute, New York University School of Medicine

Neuroscience Institute, New York University School of Medicine

Title: Oxytocin increases excitability of excitatory and inhibitory CA2 neurons through modulation of M-current

Authors: *N. N. TIRKO[1], K. W. EYRING[2], I. CARCEA[4], M. MITRE[3], M. V. CHAO[5], R. C. FROEMKE[6], R. W. TSIEN[7]
[1]Actuated Med. Inc, Warriors Mark, PA; [2]Neurosci., [3]Physiol. and Neurosci., New York Univ. Sch. of Med., New York, NY; [4]Skirball Inst., NYU Med. Ctr., New York, NY; [5]Skirball Inst. of Biomolecular Med., New York Univ. Langone Med. Ctr., New York, NY; [6]Otolaryngology, NYU Med., New York, NY; [7]Neurosci. Inst., New York Univ., New York, NY

Abstract: Oxytocin receptor (OXTR) signaling has been implicated in many aspects of social behavior, with loci throughout the mammalian brain. Consistently, one major effect of oxytocin signaling is to refine inhibitory tone, along with excitatory output. We report that within area CA2, a hippocampal subregion recently implicated in social memory, OXTR signaling stimulates both excitatory and inhibitory neurons, while sculpting output to CA1. Exogenous application of an oxytocin receptor agonist (TGOT) or optogenetic stimulation of

endogenous oxytocinergic fibers in CA2 can drive otherwise quiescent pyramidal cells to become excited. In all CA2 pyramidal neurons tested (adult mice), OXTR stimulation caused significant depolarization. This depolarization was accompanied by an increase in whole-cell membrane resistance, suggesting modulation of a potassium conductance. Indeed, TGOT-induced depolarizations were blocked by application of retigabine (100 μM), an opener of the KCNQ potassium channel responsible for the M-current. In response to TGOT application, CA2 pyramidal cells depolarize and enter into a burst-firing mode. Bursting in CA2 pyramidal cells was accompanied by a decrease in action potential amplitude and after-hyperpolarization (AHP) magnitude, facilitating high-frequency burst activity. Both phenomena rely on phospholipase C (PLC) activation, while the effects on spike and AHP magnitude depend on protein kinase C (PKC) activation. Pharmacological inhibition of the M-current and activation of PKC recapitulate the effect of OXTR stimulation in CA2 pyramidal cells. PV+ interneurons in CA2 were also depolarized by OXTR stimulation that depended on M-current inhibition. Blockade of inhibition during OXTR agonist application altered burst duration and frequency, suggesting that CA2-mediated inhibition is important in shaping the response to OXTR stimulation. Indeed, our optogenetic experiments suggest that the burst structure, shaped by CA2-mediated inhibition, is optimal for excitatory charge transfer onto CA1 pyramidal cells. We find evidence for a similar molecular mechanism at play in CA1 fast-spiking interneurons, suggesting that OXTR-mediated inhibition of M-current may be a modulatory motif throughout the brain. Further, our results suggest that oxytocinergic modulation of CA2 may impact not only CA2-dependent social memory, but more broadly, hippocampal information transfer via CA1.

Disclosures: **N.N. Tirko:** None. **K.W. Eyring:** None. **I. Carcea:** None. **M. Mitre:** None. **M.V. Chao:** None. **R.C. Froemke:** None. **R.W. Tsien:** None.

Poster

426. Learning and Memory: Hippocampal CA2 and Social Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 426.06/UU19

Topic: H.01. Animal Cognition and Behavior

Title: The contribution of hippocampal oxytocin receptors to social memory processing

Authors: *T. RAAM
Ctr. for Regenerative Med., Massachusetts Gen. Hosp., Boston, MA

Abstract: The trisynaptic hippocampal pathway DG-CA2/3-CA1 plays a critical role in processing contextual information by encoding distinct representations in non-overlapping ensembles of neurons, a process known as pattern separation. Recent studies suggest a role for the dorsal CA2/3 subregion in processing social memories, raising the question of how the same hippocampal sub-regions process social information. Based on the enrichment of receptors for

the social hormone oxytocin (Oxtrs) in the DG-CA2/3 axis, but not CA1, and the critical role oxytocin plays in social memory, we hypothesized that Oxtrs in dorsal DG-CA2/3 are essential for social memory processing. Here we employ pharmacological and genetic tools to demonstrate the necessity of Oxtrs in dDG and dCA2/3 for social memory, but not object memory. Further, we utilized ensemble mapping techniques to demonstrate that Oxtrs are necessary for population-based encoding of social stimuli in CA3. Optogenetic terminal-specific silencing revealed roles for distinct dCA2/3 outputs to downstream limbic regions in discrimination of objects and social stimuli. Together, these studies begin to elucidate how an evolutionarily conserved neuromodulatory hormone, oxytocin, utilizes a basic memory processing circuit in the hippocampus to modulate social behavior.

Disclosures: T. Raam: None.

Poster

426. Learning and Memory: Hippocampal CA2 and Social Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 426.07/UU20

Topic: H.01. Animal Cognition and Behavior

Support: NIH Intramural Research Program Z01ES100221

Title: Role of CA2 neuronal activity level in conditioned fear learning

Authors: *G. M. ALEXANDER[1], N. V. RIDDICK[2], D. J. LUSTBERG[4], S. S. MOY[3], S. M. DUDEK[5]
[1]Neurobio. Lab., Natl. Inst. of Envrn. Hlth. Sci., Research Triangle Park, NC; [2]Carolina Inst. for Developmental Disabilities and Dept. of Psychiatry,, [3]Psychiatry, Univ. of North Carolina at Chapel Hill Sch. of Med., Chapel Hill, NC; [5]Neurobio. Lab., [4]Natl. Inst. of Env. Hlth. Sci., NIH, Research Triangle Park, NC

Abstract: Hippocampal pyramidal cells (PCs) are critical for certain forms of learning and memory, and work from our lab and others has shown that PCs in CA2 are required for social cognition and behavior. Permanently silencing CA2 PCs in mice impairs social memory, and we found that chemogenetically increasing or decreasing CA2 PC activity increases or decreases sociability, respectively. Further, mice lacking Regulator of G-Protein Signaling 14 (RGS14), a protein that is highly enriched in and restricted to CA2 PCs, learn faster than wild-types (WTs) in the Morris water maze spatial memory test. CA2 PCs in these RGS14 knock-out (KO) mice have increased excitability and a capacity for LTP at CA3 □CA2 synapses in stratum radiatum, where LTP normally does not occur. Although the enhanced spatial learning abilities of the RGS14 KO mice suggest a role for CA2 PCs in at least one hippocampus-dependent behavior, the role of CA2 PCs in fear conditioning is unknown. Fear conditioning, a form of associative

learning, involves the hippocampus and extrahippocampal brain structures such as the prefrontal cortex and amygdala. Because we found that chemogenetic modification of CA2 PC excitability altered prefrontal cortical activity, we asked how modifying CA2 PC activity would affect fear conditioning. We expressed Gq- or Gi-coupled DREADDs in CA2 PCs and administered CNO before the shock-tone-context pairing. On subsequent days, we measured freezing behavior in the same context but without the tone (contextual fear) or in a new context but in the presence of the tone cue (cued fear). We found that activation of CA2 PCs during the shock pairing resulted in increased freezing behavior in male and female mice upon cue presentation. Because both Gq DREADD expression and RGS14 deletion increases CA2 PCs activity, we also measured conditioned fear responses in RGS14 KO mice. We predicted that, similar to Gq-DREADD-expressing mice, RGS14 KO mice would show enhanced freezing behavior. Indeed, RGS14 KO mice had increased freezing upon cue presentation relative to WTs. Interestingly this effect was only seen in female KO mice. Emerging evidence from our lab suggests that sex differences arise following manipulations of CA2 activity; for instance, chemogenetic inhibition of CA2 PCs during the shock pairing resulted in increased freezing in the associated *context* in female, but not male, mice. These findings support the conclusion that CA2 PC activity plays a functional role in the corticolimbic circuitry that includes prefrontal cortex and amygdala. Given the behavioral sex differences in the Gi-DREADD and RGS14KO mice, we propose that the function of CA2 PCs in this circuitry may be sexually dimorphic.

Disclosures: G.M. Alexander: None. **N.V. Riddick:** None. **D.J. Lustberg:** None. **S.S. Moy:** None. **S.M. Dudek:** None.

Poster

426. Learning and Memory: Hippocampal CA2 and Social Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 426.08/UU21

Topic: H.01. Animal Cognition and Behavior

Support: ZIA-MH-002498-24

Title: Social and non-social encoding by Vasopressin 1b receptor expressing Pyramidal neurons in CA2 hippocampal subfield of mice

Authors: *A. CYMERBLIT-SABBA[1], M. STACKMANN[1], S. K. WILLIAMS AVRAM[1], M. C. GRANOVETTER[1], A. SMITH[1], J. SONG[1], J. FASTMAN[1], H.-J. LEE[2], W. S. YOUNG[1]
[1]NIMH, Bethesda, MD; [2]Microbiology & Immunol., Kyungpook Natl. Univ., 2177 Dalgubeol-daero, Jung-gu, Daegu 41940, Korea, Republic of

Abstract: The hippocampal CA2 regions has been shown to be involved in social and contextual memory.

Still, the way these memories are encoded by it remains, for the most part, unknown.
We expressed GCaMP6s in the CA2 of vasopressin 1b receptor promoter- driven *Cre* recombinase-expressing knock-in mice and imaged the calcium transients in selective pyramidal neurons within the CA2 subfield of socially behaving mice. We used behavioral paradigms such as object and social habituation-dishabituation and social recognition discrimination to map the neural response to the stimuli presented.
Our results indicate the cells are responsive to both social and non-social stimulation. We observe an increased number of calcium events upon presentation of the novel stimulus and a subsequent decrease when the stimulus is removed from the cage. Moreover, upon repetitions of both social and non-social stimuli, neural representation becomes sparser and activity is attenuated in a stimulus-specific manner. Although the cells response to both stimuli, they response significantly more when the social stimulus is presented.
Our preliminary results suggest a preferred social encoding by these vasopressin 1b receptor-expressing pyramidal neurons.
This research was supported by the intramural research program of the NIMH (ZIA-MH-002498-24).

Disclosures: A. Cymerblit-sabba: None. **M. Stackmann:** None. **S.K. Williams Avram:** None. **M.C. Granovetter:** None. **A. Smith:** None. **J. Song:** None. **J. Fastman:** None. **H. Lee:** None. **W.S. Young:** None.

Poster

426. Learning and Memory: Hippocampal CA2 and Social Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 426.09/UU22

Topic: H.01. Animal Cognition and Behavior

Support: NIMH Grant ZIAMH002498

Title: The role of vasopressin 1b receptor-expressing pyramidal neurons of the hippocampus CA2 region in an object novelty task

Authors: *M. STACKMANN, A. CYMERBLIT-SABBA, M. C. GRANOVETTER, S. WILLIAMS, W. S. YOUNG
Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: The CA2 region of the hippocampus, unique in its synaptic mechanisms and cellular properties, has been shown to encode spatial features in the environment to a lesser extent than its neighboring CA1 or CA3 subregions. Moreover, CA2 neurons change their representation of the environment through time, or when social or novel stimuli are introduced to the environment. To further investigate the way the neurons of the CA2 encode objects in their environment, we

selectively imaged pyramidal neurons of the region by expressing the calcium indicator GCaMP6s in vasopressin 1b receptor (Avpr1b) promoter-driven Cre recombinase transgenic mice. This technique allowed us to image the calcium transients of these CA2 subfield cells as mice performed an object habituation-dishabituation paradigm. Our results suggest that CA2 pyramidal neurons are most sensitive to the initial presentation of a novel object in the environment, with sparser neural representation upon repetitions and decreased population response. Moreover, upon presentation of a second novel object, activity remains low. Our data suggest that the tuning of vasopressin 1b receptor-expressing pyramidal neurons to an object presented is invariant to the object's features.

Disclosures: M. Stackmann: None. **A. Cymerblit-Sabba:** None. **M.C. Granovetter:** None. **S. Williams:** None. **W.S. Young:** None.

Poster

426. Learning and Memory: Hippocampal CA2 and Social Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 426.10/UU23

Topic: H.01. Animal Cognition and Behavior

Support: Ville de Paris 2013 Programme Emergences

ANR-12-BSV4- 0021-01

ANR-13-JSV4-0002-01

Title: How dynamic inhibitory control of CA2 pyramidal cell output influences CA1 activity

Authors: *R. A. PISKOROWSKI[1], K. NASRALLAH[2], L. THERREAU[1], V. ROBERT[1], V. CHEVALEYRE[1]
[1]Inserm U894, Univ. Paris Descartes, Paris, France; [2]Albert Einstein Col. of Med., Bronx, NY

Abstract: The hippocampus is a critical structure for memory formation. Area CA2 has recently emerged as an important region in the control of hippocampal activity and social memory formation. However, how CA2 neurons precisely integrate in the hippocampal network is still unclear. In area CA2, the density of interneurons, and in particular, parvalbumin-expressing (PV+) interneurons is very high. Furthermore, while CA2 pyramidal cells are highly resistant to synaptic plasticity, the inhibitory transmission in this region is highly plastic, and mediated by delta opioid receptor (DOR) activation. Here, we use opto- and chemo-genetics to examine how PV+ interneuron transmission and plasticity controls CA2 pyramidal neuron input and the consequences this has on CA1. Using inhibitory DREADD expression, we found that transmission from PV+ interneurons largely contributes to the spontaneous inhibitory transmission in CA2 and to the feed-forward inhibition between CA3 and CA2. Furthermore,

silencing PV+ interneurons strongly increased the amplitude of excitatory potential from CA3. This manipulation also prevented by occlusion DOR-mediated long-term depression of inhibitory transmission (iLTD). Likewise, we found that excitatory synaptic input from the supramammillary hypothalamic input is also tightly controlled by PV+ inhibitory transmission, and this input is equally sensitive to DOR-mediated iLTD. We then investigated how plasticity in area CA2 influences CA1 activity following stimulation of CA3 inputs. We found that induction of DOR-mediated iLTD in area CA2 resulted in a significant increase in the excitatory drive onto CA1 neurons. We then used a transgenic mouse line to selectively express cre recombinase in CA2 pyramidal neurons to express in order to use optogenetics to study the connection between CA2 and CA1. We found that excitatory transmission from CA2 is larger onto deep compared to superficial CA1 neurons. We also found that CA2 pyramidal neurons strongly target an unusual group of CA1 pyramidal neurons with soma located in stratum radiatum. In addition, we found that feed-forward inhibition between CA2 and CA1 normalizes the net excitatory drive onto the deep and superficial CA1 neurons. In conclusion, we found that feed-forward inhibition mediated by PV+ interneurons controls both the input and the output of CA2 pyramidal neurons. This inhibitory transmission acts to normalize the strength of the excitatory drive between CA2 and different subclasses of CA1 pyramidal neurons.

Disclosures: R.A. Piskorowski: None. **K. Nasrallah:** None. **L. Therreau:** None. **V. Robert:** None. **V. Chevaleyre:** None.

Poster

426. Learning and Memory: Hippocampal CA2 and Social Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 426.11/UU24

Topic: H.01. Animal Cognition and Behavior

Title: Differential contributions of dorsal and ventral CA3 to social memory

Authors: *M.-C. CHIANG[1,2], A. J. Y. HUANG[1], R. BOEHRINGER[1], D. POLYGALOV[1], T. OHSHIMA[2], T. J. MCHUGH[1]
[1]RIKEN BSI, Wako City, Japan; [2]Dept. of Life Sci. and Med. Biosci., Waseda Univ., Tokyo, Japan

Abstract: Social memory is crucial for normal daily life in many animals, including rodents and primates; however, the neural circuits underlying these processes are not fully understood. Recent results have suggested an important role of the hippocampus in social memory, particularly the CA1 and CA2 subregions. Here we employ circuit genetics to address the potential contribution of the hippocampal regions upstream of CA1 and CA2, the CA3 and the dentate gyrus (DG). We applied region specific *N*-methyl-$_D$-aspartate receptor subunit 1 (NR1) knockout (KO) approaches to test the contribution of plasticity in CA3 and the DG in social

memory. We found that CA3 NR1-KO mice demonstrated an impairment of social memory, while DG NR1-KO mice were indistinguishable from controls. We next applied the designer receptors exclusively activated by designer drugs (DREADD)-based chemogenetic approach in the CA3 specific Cre-transgenic mouse to differentiate the contribution of the dorsal CA3 versus the ventral CA3 in social memory. Our results provide the evidence of differential roles of hippocampal CA3 in social memory across the dorsal-ventral axis.

Disclosures: M. Chiang: None. **A.J.Y. Huang:** None. **R. Boehringer:** None. **D. Polygalov:** None. **T. Ohshima:** None. **T.J. McHugh:** None.

Poster

426. Learning and Memory: Hippocampal CA2 and Social Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 426.12/UU25

Topic: H.01. Animal Cognition and Behavior

Support: MEXT Bionavigation Innovative Research Area

Title: The impact of transient silencing of the retrosplenial cortex on hippocampal physiology

Authors: *H. GUAN[1], A. J. Y. HUANG[2], D. POLYGALOV[1], T. INOUE[3], T. J. MCHUGH[1]
[1]BSI, RIKEN, Wako-Shi, Japan; [2]Riken BSI, Wakoshi, Japan; [3]Waseda Univ., Tokyo, Japan

Abstract: The retrosplenial cortex (RSC) is one of the major cortical outputs of the hippocampal formation and as such, has been implicated in memory formation and recall. Further, the RSC is one of the cortical structures important for spatial navigation, containing neurons responsive to the animal's trajectory and head direction. Given its close functional and anatomical connections with the CA1 region an important question is how manipulations of neuronal activity in the RSC and/or CA1 influences these processes. Here we combine transgenic mice expressing the Cre recombinase specifically in the RSC or CA1 pyramidal cells with Cre-dependent AAV vectors expression Gi-DREADD to inducibly inhibit neuronal activity in each region while monitoring in vivo physiology. We found CNO-mediated silencing of RSC activity leads to changes in hippocampal activity supporting its role in memory consolidation.

Disclosures: H. Guan: None. **A.J.Y. Huang:** None. **D. Polygalov:** None. **T. Inoue:** None. **T.J. McHugh:** None.

Poster

426. Learning and Memory: Hippocampal CA2 and Social Learning

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 426.13/UU26

Topic: H.01. Animal Cognition and Behavior

Support: JSPS Fellow

Title: Characterization of a novel hippocampal-septal circuit

Authors: *H. HE[1,2], K. OKANOYA[2], T. J. MCHUGH[1,2]
[1]RIKEN, BSI, Saitama, Japan; [2]The Univ. of Tokyo, Tokyo, Japan

Abstract: The connections between the hippocampus and medial septum (MS) are crucial for integration of sensory and motor information in memory and the generation of the theta oscillation. While recent work has uncovered specific roles for ascending cholinergic, glutamatergic and GABAergic inputs from the MS to the hippocampus in these processes, much less is known about the role of the descending hippocampal efferents. Here we use transgenic and viral methods to identify and characterize a novel descending projection pathway from the hippocampal CA2 region to the MS. We find that optogenetic manipulation of these axons alters the animals' exploratory behavior and use in vivo physiological recordings to uncover the physiological consequences of these circuit manipulations.

Disclosures: H. He: None. **K. Okanoya:** None. **T.J. McHugh:** None.

Poster

427. Hippocampal and Cortical Circuits Mediating Learning and Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 427.01/UU27

Topic: H.01. Animal Cognition and Behavior

Support: NIH R21 DC015602

Title: Head direction cells in mice are referenced to gravity

Authors: *H. CHAM[1], J. LAURENS[1], D. E. ANGELAKI[2], J. DICKMAN[3]
[1]Neurosci., [2]Dept of Neurosci., [3]Dept. of Neurosci., Baylor Col. of Med., Houston, TX

Abstract: Head direction cells (HDC) form a “neural compass” of the local environment and play an important role in spatial navigation. Recent evidence reported that head direction cells in bats were 3D spatially tuned, suggesting that their neural response was modulated by the tilt relative to gravity (Finkelstein et al., 2015). Furthermore, we have identified gravity-referenced tilt signals in the anterior thalamus (ATh) of macaques (Laurens et al., 2016). However, ATh neurons in our previous study were not specifically identified as HDC. Furthermore, it remains unknown whether these navigation circuit neurons encode a 3D neural compass. Here, we examined whether HDC in the ATh of mice encode tilt information and determine whether the tilt encoding is referenced to gravity.

We recorded the response of ATh head direction cells in mice (n = 6) by manipulating the restrained animals’ 3D head orientation using a multi-axis motion stimulator. Neural activity was recorded using a chronic tetrode implant. Head direction responses were measured separately when animals moved freely in a circular arena. We recorded the response of 153 cells, out of which 32 (21%) were identified as HDC and 121 (79%) were non-HDC.

The majority of ATh cells (97/153, 63%) had a preferred direction of tilt orientation. In addition, both HD cells (21/32, 66%) and non-head direction cells (76/121, 63%) responded to tilt (Chi Square: p = 0.9). We then examined whether the tilt encoding was referenced either visual or gravity cues. First, we found that the tuning curves of these neurons were similar in both the light and the dark in terms of: preferred direction (r = .60, $p < 10^{-3}$), tuning width (r = .42, $p < 10^{-3}$), minimal firing rate (r = .95, $p < 10^{-5}$), and maximum firing rate (r = .96, $p < 10^{-5}$). Next, we measured the neural response tuning after placing the visual and gravity reference frames in conflict by tilting the visual surround while gravity remained constant. We found that cell responses were better correlated when computed in a gravity reference frame as compared the visual reference frame (Wilcoxon signed rank: $p < 10^{-4}$).

These results are the first to demonstrate tilt encoding of HDC in a land-dwelling animal. Our data suggests that tilt-sensitive ATh neurons use the gravity signal as a reference. The finding that tilt encoding is referenced to gravity highlights the role of the vestibular system in establishing self-orientation during navigation.

Disclosures: **H. Cham:** None. **J. Laurens:** None. **D.E. Angelaki:** None. **J. Dickman:** None.

Poster

427. Hippocampal and Cortical Circuits Mediating Learning and Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 427.02/UU28

Topic: H.01. Animal Cognition and Behavior

Support: NIH R21 DC015602

Title: Neural encoding of azimuth by head direction cells during rotation in three dimensions; and interaction with gravity responses

Authors: J. LAURENS[1], H. CHAM[1], *J. DICKMAN[2], D. E. ANGELAKI[3]
[1]Neurosci., [2]Dept. of Neurosci., [3]Dept of Neurosci., Baylor Col. of Med., Houston, TX

Abstract: Here, we examined whether head direction cells (HD) in the Anterior Thalamus (ATh) of mice encode a 3D neural compass. We recorded the responses of HD cells across a broad range of 3D head orientations during continuous motion stimulation in 2D tilt and 1D azimuth. We observed that the 3D HD responses can be decomposed into: (1) a 'gravity' component during head tilt in vertical planes previously observed in bats, primates and rodents (Finkelstein et al. 2015, Laurens et al 2016, and companion poster: Cham et al. 2017), and (2) an 'azimuth' component where HD encode head orientation in the horizontal plane when animals are upright. Previous studies have suggested that the azimuth plane may follow an animal's plane of locomotion when walking on vertical surfaces (Calton et al 2005; Wilson et al. 2016) or that azimuth and pitch may form a toroidal geometry (Finkelstein et al. 2015). However, a general understanding of neural encoding of azimuth though the whole 3D orientation space has yet to be described.

We recorded the activity of 99 ATh neurons during a passive rotation protocol across the whole 3D orientation space. We observed that 35/99 neurons had significant ($p<0.05$) azimuth tuning during this passive rotation protocol and were classified as HD cells. Nineteen of the thirty five neurons also exhibited a significant gravity tuning, and an additional 34/99 cells were tuned to gravity but no azimuth.

We observed, in agreement with Finkelstein et al. 2015 in bat HD cells, that the azimuth preferred directions (PD) reversed when animals were pitched beyond 90° upward or downward (but not rolled). In contrast, the PD for these cells didn't reverse when animals were rolled left or right up to 150°. We observed intermediate PD shifts for 90° to 150° tilt around intermediate (pitch/roll) axes. Azimuth tuning in upside-down orientation (>150° pitch or roll) was weak and variable. We propose a geometrical definition of azimuth during 3D rotation that accounts for these results and is compatible with the topologies proposed by Finkelstein et al. 2015 and Wilson et al. 2016.

We also found the same pattern of azimuth tuning in a subgroup of 19 gravity-tuned HD cells. The azimuth and gravity tunings interacted in these cells to form a preferred 3D orientation, which we described and modeled . Finally, we verified that this pattern of 3D orientation tuning uncovered during passive movements of the head-fixed animals persists during active exploration in 3D space in unrestrained freely-moving mice.

Disclosures: J. Laurens: None. **H. Cham:** None. **J. Dickman:** None. **D.E. Angelaki:** None.

Poster

427. Hippocampal and Cortical Circuits Mediating Learning and Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 427.03/UU29

Topic: H.01. Animal Cognition and Behavior

Support: R01 MH102394

Title: Comparison of medial entorhinal and nucleus reuniens projections in ca1

Authors: *D. MAISSON, A. L. GRIFFIN
Psychological and Brain Sci., Univ. of Delaware, Newark, DE

Abstract: Our lab has demonstrated that the ventral midline thalamic nucleus reuniens (Re) plays a vital role in spatial working memory (SWM) and oscillatory synchrony between the dorsal hippocampal (HPC) CA1 sub-region and the medial prefrontal cortex (mPFC) (Hallock et al., 2016), likely due to its extensive anatomical connectivity with both structures. Previous work has demonstrated that stratum lacunosum-moleculare (SLM) of CA1 receives dense projections from both Re and MEC. Considering these findings and that these pathways are both critical for SWM, the current study aims to characterize the shared and differential points of synaptic contact of the projections from each structure. This study used two distinct adeno associated viral vectors, each tagged with a different fluorophore, to compare the extent of overlapping innervation from Re and MEC to CA1. Within a given rat, a viral vector tagged with GFP was injected into the Re and one tagged with tdTomato was injected into the MEC. We allotted 6 weeks for expression of the fluorophore along the extent of the membrane and imaged the terminals in CA1 using confocal microscopy at multiple magnifications. Consistent with previous findings, our preliminary data show that stratum SLM receives projections from both the Re and the MEC. Interestingly, within SLM, we found expression of terminals from both structures in close proximity, which is suggestive of dual innervation from Re and MEC onto the same neuronal compartments. Future work will be aimed at identifying whether these two projection pathways converge on dendritic spines of pyramidal neurons, inhibitory interneuron dendrites, or both.

Disclosures: D. Maisson: None. **A.L. Griffin:** None.

Poster

427. Hippocampal and Cortical Circuits Mediating Learning and Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 427.04/UU30

Topic: H.01. Animal Cognition and Behavior

Support: R01 MH102394

Title: Distinct spatial working memory correlates of prefrontal and hippocampal projections to the nucleus reuniens

Authors: *Z. M. GEMZIK, J. J. STOUT, M. T. GAYLORD, D. MAISSON, A. L. GRIFFIN
Psychological and Brain Sci., Univ. of Delaware, Newark, DE

Abstract: Spatial working memory (SWM) requires the encoding, maintenance and retrieval of spatially relevant information that is then used to successfully navigate toward a goal and relies both on the medial prefrontal cortex (mPFC) and hippocampus (HPC). Due to reciprocal mPFC and HPC connections, the ventral midline thalamic nucleus reuniens (Re) is ideally situated to orchestrate mPFC-HPC interactions. Indeed, our lab has shown that suppression of Re cell body activity is critical for mPFC-HPC synchrony and SWM (Hallock et al., 2016). However, it is not yet clear what distinct roles the specific inputs into the Re have on particular phases of SWM. In order to address this question, we used archaerhodopsin (pAAV5-CAG-ArchT-GFP) for temporally selective optogenetic terminal suppression of either mPFC or dorsal subiculum (dSub) projections to the Re during either the cue-guided, memory-guided, or delay/maintenance phase of the Delayed Non-Match to Position (DNMP) task. Rats were injected with virus in either the mPFC or the HPC; for all rats, the fiber was implanted into the Re. This approach allowed us to selectively suppress terminals from either structure to the Re during each phase of DNMP (sample, choice, delay, and entire-trial). Our data show a significant impairment when mPFC terminals were suppressed during the choice phase but not sample, delay, or entire-trial phases of the task. Conversely, suppression of dSub terminals led to a trending deficit exclusively during the sample phase. These results suggest that the mechanisms underlying encoding may be supported by the HPC-Re-mPFC pathway whereas retrieval may be supported by the and mPFC-Re-HPC pathway.

Disclosures: Z.M. Gemzik: None. **J.J. Stout:** None. **M.T. Gaylord:** None. **D. Maisson:** None. **A.L. Griffin:** None.

Poster

427. Hippocampal and Cortical Circuits Mediating Learning and Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 427.05/UU31

Topic: H.01. Animal Cognition and Behavior

Support: R01 MH102394

Title: Oscillatory synchrony within the hippocampal-thalamo-prefrontal circuit during spatial working memory

Authors: *A. C. GARCIA, A. L. GRIFFIN
Univ. of Delaware, Newark, DE

Abstract: Spatial working memory-guided behavior in rodents has been shown to be supported by oscillatory synchrony between the hippocampus and the medial prefrontal cortex (mPFC). Extracellular recordings of local field potentials in dorsal hippocampus and mPFC have shown increased coherence in the theta band (4-12 Hz) when animals are required to apply spatial working memory (WM) toward a decision-making process. However, anatomical connections between hippocampus and mPFC are not bi-directional and are limited to a monosynaptic pathway extending from hippocampus to mPFC, primarily via its ventral subregion. The nucleus reuniens (RE) of ventral-midline thalamus is not only reciprocally connected with both hippocampus and mPFC, but has also been shown to be necessary for spatial WM-guided behavior. Furthermore, pharmacological inactivation of RE during a delayed-alternation task on a T-maze led to concomitant disruptions in theta coherence, theta-gamma phase-amplitude coupling, and directionality between dorsal hippocampus and mPFC when rats were required to emit a spatial WM-guided response (Hallock et al., 2016). To begin to understand how RE participates in hippocampal-mPFC interactions during spatial WM, we recorded local field potentials in dorsal hippocampus, RE, and mPFC during the delayed non-match to position task (DNMP) on a T-maze. The DNMP task requires encoding trial-specific information during the sample phase, maintenance across a delay period, and retrieval-guided decision making during the choice phase. We hypothesized that inter-regional oscillatory synchrony would be increased for the choice phase compared to the sample phase. In line with previous reports, we observed higher hippocampal-mPFC theta coherence during the choice phase compared to the sample phase (O'Neill et al., 2013). Additionally, coherence between RE and mPFC showed increases in both the theta frequency and the slower delta frequency (2-5 Hz) for the choice phase compared to the sample phase. Furthermore, these choice phase-specific increases in synchrony were not observed when animals performed the task poorly. Although coherence between RE and hippocampus showed peaks at theta, we did not observe any differences between sample and choice. The delta/theta coherence between RE and mPFC during the choice phase may serve to

co-modulate ensembles in mPFC that support spatial WM representations (Fujisawa et al., 2011). Further analyses will examine and characterize cross-frequency coupling and directionality within this tri-regional circuit during spatial WM.

Disclosures: A.C. Garcia: None. **A.L. Griffin:** None.

Poster

427. Hippocampal and Cortical Circuits Mediating Learning and Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 427.06/UU32

Topic: H.01. Animal Cognition and Behavior

Support: NSF GRF DG-1144086

NSF IOS-1149718

KIBM IRG 2014-004

Title: Posterior parietal and retrosplenial cortices map the position of a pursuit target

Authors: *D. A. NITZ[1], A. S. ALEXANDER[3], J. TUNG[2]
[2]Cognitive Sci., [1]Univ. of California San Diego, La Jolla, CA; [3]Boston Univ., Boston, MA

Abstract: Rats in the wild travel in packs and hunt small prey, activities that require pursuit of a moving target. In the laboratory, our previous work has shown that rats in an open arena will pursue a laser target moving in random trajectories. Rats also execute shortcut trajectories during stereotyped laser paths interspersed amongst the random paths. The present work examined neural activity in the posterior parietal cortex (PPC) and retrospenial cortex (RSC) that registered the position of the moving target relative to the animal's head during active pursuit. Data indicate that, after controlling for the PPC and RSC correlates for the animal's movement (linear and angular velocity), neurons in the PPC and RSC encode the position of the laser target relative to the animal, at least within a 40 cm long by 60 cm wide spatial window extending out from the rat's head. Sub-populations of neurons in both regions encoded the distance of the laser target irrespective of angular offset, or exhibited laser target receptive fields that were biased according to the distance and angle of the laser target relative to the animal's head. We conclude that both RSC and PPC can effectively generate a mapping of target position that is integrated within an encoding of the animal's current linear and angular velocity.

Disclosures: D.A. Nitz: None. **A.S. Alexander:** None. **J. Tung:** None.

Poster

427. Hippocampal and Cortical Circuits Mediating Learning and Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 427.07/UU33

Topic: H.01. Animal Cognition and Behavior

Support: NSF IOS-1149718

NSF GRF DGE-1144086

Title: Representation of navigational distance in the retrosplenial cortex

Authors: *A. S. ALEXANDER[1], D. A. NITZ[2]
[1]Boston Univ., Boston, MA; [2]Univ. of California San Diego, La Jolla, CA

Abstract: Recent work has demonstrated that retrosplenial cortex (RSC) neural ensembles encode a multitude of spatial variables relevant to effective navigation, including: location and heading orientation in the environment, position within known routes, and the current or upcoming action state of the animal. However, accurate spatial mapping and movement also likely requires a representation of the animal's distance from fixed environmental points or landmarks. Recent work in humans traversing virtual environments indicates that BOLD activation in RSC can track distance information. Distance encoding likely requires neural circuitry sensitive to external location, heading, and egocentric relationship to other environmental locations or landmarks. The confluence of projections that reach RSC would potentially produce neurons simultaneously sensitive to these spatial variables, as RSC is reciprocally connected to sub-cortical structures known to represent spatial location and heading in the external world and cortical regions that process sensory information such as the positions of visual stimuli relative to the animal. Here we report that RSC ensembles encode the animal's distance from fixed points in well-known routes. This form of spatial representation is constructed from spatial activation profiles of individual RSC neurons that exhibit symmetrical patterns around individual points within the trajectory. Aligned symmetrical firing rate profiles could be utilized to accurately decode the animal's distance from these points of symmetry, effectively yielding a metric of the animal's distance from all points in the environment. We observed this form of spatial representation across multiple routes and explored distance encoding in RSC during free exploration. The current data indicates that RSC neurons generate a unique representation of navigational distance, including distance from a trajectory start point. Thus, this property is in line with deficits to path integration and topographic disorientation following damage to the region.

Disclosures: A.S. Alexander: None. **D.A. Nitz:** None.

Poster

427. Hippocampal and Cortical Circuits Mediating Learning and Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 427.08/UU34

Topic: H.01. Animal Cognition and Behavior

Support: Frontiers of Innovation Scholars Program (FISP) Fellowship, UCSD, 2016

Title: Creating spaces: Posterior parietal cortex and hippocampal mapping of environmental subspaces

Authors: *L. E. SHELLEY, D. A. NITZ
UCSD, La Jolla, CA

Abstract: The present study examined posterior parietal cortex and hippocampal representations of an environment divided into subspaces. While numerous studies have outlined posterior parietal cortex route mapping as well as hippocampal place mapping in various situations, the current study examined environmental subspace representations using a task-defined, behavioral rule to demarcate the division of a room into two subspaces, rather than a concrete feature or visible dividing line. During initial training, rats traversed a T-shaped track that was placed in one of four locations. Each rat learned to run up the stem of the 'T' and either turn left or right depending on the location of the track's choice point in the room. Rats were trained to turn left when the track was placed in one half of the room, and to turn right when placed in the other half. This rule divided the overall room into two subspaces in the absence of any physical boundary line. Behavioral results suggested that rats are capable of learning such logical fragmentation, as evidenced by high accuracy during probe trials in which the track was placed in novel locations. Furthermore, although performance in all track positions was significantly above chance, accuracy was dependent on the track's proximity to the fragment boundary, as shown by worse performance for track locations near the boundary. This was likely due to increased ambiguity in turn choice with less spatial separation between a track location requiring a left turn and one requiring a right turn. After rats were trained, neurons in both the hippocampus and posterior parietal cortex were recorded during task performance. Spatial firing patterns of neurons in both brain regions followed the track space in each of the four positions, an effect that was expected for posterior parietal cortex neurons, but was unexpected in the hippocampus. Furthermore, posterior parietal cortex ensembles discriminated between the animal's ultimate left vs right turning behavior at earlier stages in each trial than did the hippocampal population firing patterns. Findings from the present study suggest that rats are capable of learning a spatial division into subspaces that is rule-induced, and that posterior parietal cortex and hippocampal neurons modulate their typical firing patterns in a novel way to map environmental subspaces.

Disclosures: **L.E. Shelley:** None. **D.A. Nitz:** None.

Poster

427. Hippocampal and Cortical Circuits Mediating Learning and Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 427.09/UU35

Topic: H.01. Animal Cognition and Behavior

Support: NIH grant R01 ES015295

Title: Enriched environment alters the epigenetic profile in the hippocampus and mitigates memory deficits induced by early postnatal lead exposure in a sex-dependent manner

Authors: *V. SINGH, G. VARMA, J. SCHNEIDER
Anatomy, Pathology and Cell Biol., Thomas Jefferson Univ., Philadelphia, PA

Abstract: Being raised in an enriched environment improves cognitive functions and presents a potential social intervention strategy to mitigate cognitive/behavioral deficits associated with developmental disorders. However the molecular mechanisms underlying the structural and functional effects on the brain induced by an enriched environment remain poorly understood. Developmental exposure to the environmental toxicant lead (Pb) results in behavioral and cognitive impairments that can persist lifelong, both in humans and animal models. No current therapies have been effective in reversing the neurological damage and functional deficits induced by Pb exposure. The purpose of this study was to investigate the extent to which environmental enrichment could mitigate memory consolidation/recall deficits and further modify altered epigenetic marks in response to developmental Pb exposure. Following an early postnatal exposure (EPN) to Pb (birth through weaning), male and female Long Evans rats were randomly housed under non-enriched (standard housing, 3 rats/cage) or enriched (large enclosures, 6 rats/cage, equipped with variety of toys, running wheels etc., changed 3 times/week) conditions. Non Pb-exposed controls were housed under similar conditions. At PND55, the animals were tested for associative memory deficits using a trace fear conditioning paradigm. Memory deficits were observed only in non-enriched EPN females. Enriched EPN females performed similar to non Pb controls suggesting that living in the enriched environment mitigated the cognitive deficit induced by Pb exposure. To gain insight into possible epigenetic mechanisms underlying these results, we examined global DNA methylation (mC) and hydroxymethylation (hmC) levels in the CA1 region of the hippocampus. Non-enriched EPN females had globally reduced mC and hmC levels. Enriched EPN females and controls had similar mC and hmC levels and higher than those in non-enriched EPN females. Exposure to Pb differentially altered expression levels of DNA methylation-related genes in EPN male and females and environmental enrichment tended to normalize these levels, making them

comparable to controls. Further studies aimed at understanding gene level functional networks/mechanisms and their functional significance are in progress.

Disclosures: V. Singh: None. **G. Varma:** None. **J. Schneider:** None.

Poster

427. Hippocampal and Cortical Circuits Mediating Learning and Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 427.10/UU36

Topic: H.01. Animal Cognition and Behavior

Support: NINDS NS-053907

Title: Grid cell representation across a multi-level maze

Authors: *P. A. LACHANCE[1], S. S. WINTER[2], M. L. MEHLMAN[1], J. S. TAUBE[1]
[1]Psychological and Brain Sci., Dartmouth Col., Hanover, NH; [2]The Jackson Lab., Bar Harbor, ME

Abstract: Successful navigation in the real world requires understanding of three-dimensional (3D) space, though most navigation research focuses on exploration in two-dimensional (2D) arenas. Evidence for neural processing of 3D space can be found in studies examining head direction (HD) cells, which encode the instantaneous orientation of an animal's head. HD cells in the rat translate their 2D representations in accordance with movement from a horizontal to a vertical plane of locomotion. Bat HD cells show firing rate modulation in response to head rotations about the yaw, pitch, and roll axes. Studies have also been undertaken to examine 3D representations in grid cells, which represent 2D space with periodic firing fields that form a repeating hexagonal pattern across an arena. However, 3D grid cell studies have used arenas that force an animal to change its vertical position either continuously or in tandem with a change in horizontal position. The current study sought to evaluate grid representations as female rats explored the same horizontal arena at different vertical heights. Grid cells in the medial entorhinal cortex were monitored as a rat explored a gray square arena (baseline: 1.2 x 1.2 x 0.5 m). Following this, a slightly smaller arena with clear plastic walls was placed into the larger arena, and the rat was allowed to explore this environment within the smaller arena (lower: 1 x 1 x 0.5 m). For the third session, the clear-walled arena was lifted 0.5 m above the floor (upper: 1 x 1 x 0.5 m), such that the rat could look out and see the baseline arena below while still exploring within a familiar but now vertically displaced arena. Grid cells appeared to maintain the size, spacing, and orientation of nodes across the lower and upper locations. However, the grid phase was offset between sessions, such that there was no overlap between lower and upper level grid nodes. This finding indicates that grid cells might integrate vertical displacement into their spatial representations, resulting in a 3D grid pattern transected by the two horizontal planes

explored in this study. Alternatively, the rats could have perceived the upper level as a novel context, resulting in the observed shift in grid patterns.

Disclosures: P.A. Lachance: None. **S.S. Winter:** None. **M.L. Mehlman:** None. **J.S. Taube:** None.

Poster

427. Hippocampal and Cortical Circuits Mediating Learning and Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 427.11/UU37

Topic: H.01. Animal Cognition and Behavior

Support: NIH NS-053907

Title: Are head direction cell responses commutative on a 3D surface?

Authors: *J. R. DUMONT[1], P. A. LACHANCE[1], J. L. MARCROFT[1], N. R. BOVIO[1], S. S. WINTER[2], J. S. TAUBE[1]
[1]Psychological & Brain Sci., Dartmouth Col., Hanover, NH; [2]The Jackson Lab., Bar Habor, ME

Abstract: An animal's perceived sense of orientation depends upon the head direction (HD) system, which is comprised of HD cells that discharge when an animal faces a particular direction in the environment; i.e., the preferred firing direction (PFD). While most research has focused on the neural properties of HD cells in the horizontal plane, animals navigate through 3D terrains. Previous research examining HD cell responses in 3D has lead to two competing models in predicting how HD cells respond in different planes: 1) the 'rotational plane model', which postulates that as a rat moves from a horizontal surface onto a vertical one, the cell will transform its frame of reference to its current plane of locomotion by rotating the horizontal plane by 90° into the vertical plane. Thus, a cell's PFD in the vertical plane can be treated as a rotational extension of the floor, where the animal shifts its reference frame to align with its current plane of locomotion; 2) the 'mosaic model', which is based on recent findings showing that as rats traverse across vertical corners of a cuboidal structure, HD cells shift their PFDs by 90° across vertical boundaries (90° CCW shift for leftward crossings and 90° CW shift for rightward crossings). PFD shifts did not occur across horizontal boundaries. Both these models make different predictions about how HD cells would respond once they reach the top (horizontal) surface following routes across vertical boundaries. The rotational plane model predicts that if an animal always rotates its plane of locomotion (along with the cell's PFD) onto the next surface, then the cell will fire in a different direction compared with the floor when the animal reaches the top surface - depending upon the path taken (i.e., firing is not commutative). In contrast, the mosaic model predicts that a cell's PFD would remain the same when the rat travels from the floor onto a vertical surface and then across a second vertical surface before

reaching the top surface (i.e., firing is commutative) because the model argues that cells' PFDs will shift by 90° around the vertical corner, but not around the horizontal corner. To test whether cell responses are commutative, we recorded HD cells from the anterodorsal thalamus while rats traveled different paths to reach the top surface of a cube (floor -> wall -> top vs. floor -> wall -> side wall -> top). Our results indicate that the majority of HD cells shifted their PFDs by 90° around vertical corners such that the cells responses on the top surface remained similar to that on the floor regardless of the path taken by the rats - thus demonstrating commutativity. These results are more consistent with the mosaic model than the rotational plane model.

Disclosures: J.R. Dumont: None. **P.A. Lachance:** None. **J.L. Marcroft:** None. **N.R. Bovio:** None. **S.S. Winter:** None. **J.S. Taube:** None.

Poster

427. Hippocampal and Cortical Circuits Mediating Learning and Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 427.12/UU38

Topic: H.01. Animal Cognition and Behavior

Support: NS053907

NS096888

Title: The dorsal striatum and medial precentral cortex are functionally and anatomically integrated into the head direction cell circuitry

Authors: *M. L. MEHLMAN[1], S. S. WINTER[2], S. VALERIO[3], J. S. TAUBE[1]
[1]Dartmouth Col., Hanover, NH; [2]The Jackson Lab., Mount Desert, ME; [3]Natl. Inst. of Hlth. and Med. Res., Bordeaux, France

Abstract: Head direction (HD) cells provide a sense of direction used for navigation. These neurons fire as a function of the animal's HD and are located in multiple, interconnected brain regions in the limbic system. Interestingly, HD cell activity has been observed in regions outside of the canonical limbic HD circuit, including the dorsal striatum (DS) and medial precentral cortex (PrCM). We performed a series of electrophysiological and neuroanatomical studies in rats to examine the functional and anatomical relationships between the limbic HD circuit, DS and PrCM.
We first recorded HD cells in the DS and PrCM. These extralimbic HD cells were nearly identical to limbic HD cells recorded in the anterodorsal thalamic nuclei (ADN) in regards to their basic firing characteristics and responses to environmental manipulations. Angular head velocity (AHV) cells – neurons modulated by head motion – were also recorded in the DS and PrCM. We then recorded DS and PrCM activity in animals that had received ADN lesions to

disrupt the limbic HD circuit. Complete lesions abolished extralimbic HD cell activity, indicating that the HD signal in the DS and PrCM is dependent upon output from the limbic HD circuit. AHV cell activity was spared in lesioned animals, suggesting that it is either generated locally or inherited from an external structure independently of the ADN. Finally, to determine if the DS and PrCM provide self-motion information to the limbic HD circuit (e.g. a motor efference signal or AHV signal), we recorded ADN HD cells in animals with combined DS and PrCM lesions. These lesions did not affect the limbic HD signal, suggesting that self-motion information is not conveyed from the DS and PrCM to the limbic HD circuit.
Next, we performed neuroanatomical tracing experiments to identify pathways that could facilitate communication between the limbic HD circuit, DS and PrCM. Retrograde tracing revealed that the DS receives direct input from multiple regions in the limbic HD circuit, including the lateral mammillary nuclei (LMN), ADN, retrosplenial cortex (RSC) and medial entorhinal cortex (MEC). These projections preferentially target the same DS subregion in which we recorded HD cells and AHV cells. The PrCM receives direct input from the LMN, RSC and MEC. Direct projections from the LMN to the DS and PrCM may convey AHV information independently of the ADN. Anterograde tracing revealed that the DS and PrCM indirectly project to the limbic HD circuit via the entopeduncular nucleus.
Collectively, these foundational studies describe how the DS and PrCM are functionally and anatomically integrated into the HD cell circuitry, expanding our understanding of the neural basis of navigation.

Disclosures: **M.L. Mehlman:** None. **S.S. Winter:** None. **S. Valerio:** None. **J.S. Taube:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.01/UU39

Topic: H.01. Animal Cognition and Behavior

Support: Shota Rustaveli National Science Foundation, Grant №3/10

Title: Modulation of GluN2B subunit-containing NMDA receptors expression and spatial long-term memory in medial septal immunolesioned rats

Authors: *G. BESELIA, M. DASHNIANI, M. BURJANADZE, R. SOLOMONIA, L. KRUASHVILI, N. CHKHIKVISHVILI
Behavior and Cognitive Functions, I. Beritashvili Ctr. of Exptl. Biomedicine, Tbilisi, Georgia

Abstract: The hippocampus is important in the formation of spatial memory in both humans and animals. The N-methyl-D-aspartate (NMDA) type of glutamate receptors in the hippocampus has been reported to be essential for spatial learning and memory as well as for the induction of

synaptic plasticity. Evidence accumulated from recent studies suggest that GluN2A and GluN2B subunit-containing NMDA-Rs preferentially contribute to the induction of hippocampal LTP and LTD. Using a Morris water maze (MWM) task, the LTP-blocking GluN2A antagonist had no significant effect on any aspect of performance, whereas the LTD-blocking GluN2B antagonist impaired spatial memory consolidation.
The present study was designed to investigate the effect of selective immunolesions of cholinergic and GABA-ergic septohippocampal projection neurons [using 192 IgG-saporin (SAP) or GAT1-1 saporin (GAT), respectively] on spatial memory assessed in MWM and NMDA receptor GluN2B subunit expression in the rat hippocampus. We used MWM training protocol with eight training trials. One day after training, probe test with the platform removed was performed to examine long-term spatial memory retrieval. We found that immunolesions of medial septal cholinergic or GABAergic neurons did not affect spatial learning as exhibited by a decreased latency to find the hidden platform across the eight training trials. Trained control and SAP treated rats spent significantly longer than chance (15 s) performances such as swimming time in test sector (where the hidden platform was located). Moreover, they spent significantly longer in test sector than the opposite sector, confirming the establishment of long-term memory. In contrast, the preference for test sector was abolished in medial septal GAT treated rats. Because GAT treated rats learned the location of the hidden platform during training, the result suggest that GAT level of NR2B subunit of NMDA receptor in the hippocampus was decreased significantly in the GAT treated group compared with the control and SAP treated groups.
In conclusion, our findings suggest that immunolesion of medial septal GABAergic neurons can interrupt hippocampus dependent spatial memory, possible through modulation of NMDA receptor subunit expression in the hippocampus. Moreover, our finding that selective lesions of medial septal GABAergic neurons affect probe-test performance but not spatial learning, suggests that septohippocampal GABAergic projections are involved specifically in the consolidation or retrieval, but not in the acquisition of long-term memory.

Disclosures: **G. Beselia:** None. **M. Dashniani:** None. **M. Burjanadze:** None. **R. Solomonia:** None. **L. Kruashvili:** None. **N. Chkhikvishvili:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.02/UU40

Topic: H.01. Animal Cognition and Behavior

Support: NSFC Grant 81630031

NSFC Grant 81471369

NSFC Grant 81571321

NSFC Grant 81571312

NSFC Grant 81401129

973 program 2015CB856400

National Key Technology R&D Program 2015BAI13B01

Title: Calbindin in distinct populations of hippocampal neurons modulates stress susceptibility and resilience

Authors: *J.-T. LI[1], X.-M. XIE[1], J.-Y. YU[2], Y.-A. SU[1], M. V. SCHMIDT[3], X.-D. WANG[2], T.-M. SI[1]
[1]Inst. of Mental Health, Peking Univ., Beijing, China; [2]Inst. of Neuroscience, Zhejiang Univ., Zhejiang, China; [3]Max Planck Inst. of Psychiatry, Munich, Germany

Abstract: Calbindin critically modulates intracellular Ca2+ dynamics and synaptic plasticity. Reductions of hippocampal calbindin levels have been implicated in cognitive disorders, including those induced by early-life stress. However, it is unclear how early-life stress modulates calbindin expression in distinct hippocampal neurons and the contribution of calbindin in each neuronal population to cognition. Here, we report that calbindin in hippocampal excitatory and inhibitory neurons modulates the susceptibility and resilience to stress-induced spatial memory deficits respectively. Stress exposure during early postnatal period lastingly reduced calbindin levels in all CA1 and DG neurons. Reduced calbindin levels in CA1 or DG excitatory neurons, but not CA1 interneurons, correlated with spatial memory impairments. Accordingly, selective knockdown of calbindin in CA1 or DG excitatory neurons mimicked early-life stress-induced memory deficits in adulthood. By contrast, calbindin knockdown in CA1 interneurons preserved memory both under basal conditions and after an acute stress challenge. Moreover, calbindin expression levels were suppressed by early-life stress through the CRHR1-nectin3 pathway, and in turn reduced inositol monophosphatase levels. Our findings highlight calbindin as a key molecule for the reprogramming effects of early-life stress on cognition, and exemplify how distinct neurons sharing a same molecule confer the susceptibility or resilience to stress.

Disclosures: **J. Li:** None. **X. Xie:** None. **J. Yu:** None. **Y. Su:** None. **M.V. Schmidt:** None. **X. Wang:** None. **T. Si:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.03/UU41

Topic: H.01. Animal Cognition and Behavior

Support: NIH Grant P20GM0103423

Pitt Hopkins Research Foundation

Title: Targeting DNA methylation to enhance learning and memory

Authors: K. E. ZENGELER, C. P. GETTENS, A. R. GOGLIETTINO, T. S. R. HOTZ, B. G. MALACHOWSKY, *A. J. KENNEDY
Chem., Bates Col., Lewiston, ME

Abstract: The storage and retrieval of memories is driven by a series of biochemical reactions that ultimately alter the expression of plasticity-regulating genes in participating neurons. Epigenetic mechanisms, such as DNA methylation, can regulate levels of gene expression and have been associated with altered plasticity in memory circuits. Specifically, active DNA methylation has been demonstrated to be necessary for the formation and consolidation of long-term memory. Here, we investigated the TET family of dioxygenase enzymes that drive active cytosine DNA demethylation in the CNS as targets for enhancing memory formation and storage. By the selective knock out or ASO-targeted knock down of the different isoforms of the TET family of enzymes, we examined isoform efficacy at altering the methylation and expression of plasticity-related genes in the hippocampus. Next, we determined whether these TET-driven changes were sufficient to enhance spatial and context-dependent long-term memory. Finally, we examined whether targeting TET isoforms is sufficient to treat the cognitive deficits associated with Pitt Hopkins Syndrome (PTHS), a monogenetic autism-spectrum disorder caused by haploinsufficiency of *Tcf4* that is associated with reduced DNA methylation in the hippocampus. Altogether, our results indicate a therapeutic potential for TET enzyme inhibition to improve learning and memory in diseases and disorders of memory.

Disclosures: K.E. Zengeler: None. **C.P. Gettens:** None. **A.R. Goglietino:** None. **T.S.R. Hotz:** None. **B.G. Malachowsky:** None. **A.J. Kennedy:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.04/UU42

Topic: H.01. Animal Cognition and Behavior

Support: NRF Grant 2012R1A3A1050385

NRF Grant 2016R1D1A1B03931525

Title: LSD1 phosphorylation by PKCα is required for presynaptic plasticity and hippocampal learning and memory

Authors: *C.-S. LIM[1], H. NAM[2], J. LEE[1], D. KIM[2], J. CHOI[1], S. J. KANG[1], S. KIM[1], H. KIM[1], C. KWAK[1], K.-W. SHIM[1], S. KIM[1], H.-G. KO[1], R. LEE[1], E.-H. JANG[1], J. YOO[1], J. SHIM[1], M. ISLAM[1], Y.-S. LEE[3], J.-H. LEE[4], S. BAEK[2], B.-K. KAANG[1]
[1]Lab. of Neurobiology, Seoul Natl. Univ., Seoul, Korea, Republic of; [2]Lab. of Mol. and Cell. Genetics, Seoul Natl. Univ., Seoul, Korea, Republic of; [3]Dept. of Physiol., Seoul Natl. Univ. Col. of Med., Seoul, Korea, Republic of; [4]Dept. of Life and Nanopharmaceutical Sci., Kyung Hee Univ., Seoul, Korea, Republic of

Abstract: The epigenetic changes by histone modification is important for synaptic plasticity and brain function. Lysine-specific demethylase 1 (LSD1) is a histone demethylase that participates in transcriptional repression or activation. Recent studies reported that LSD1 is involved in learning and memory. Although LSD1 phosphorylation by PKCα was implicated in circadian rhythmicity, the importance of LSD1 phosphorylation by PKCα in learning and memory is unknown. In this study, we examined the roles of LSD1 in synaptic plasticity and memory using *Lsd1*$^{SA/SA}$ knock-in (KI) mice, in which a PKCα phosphorylation site of LSD1 is mutated. Interestingly, short-term and long-term contextual fear memory as well as spatial learning and memory were impaired in *Lsd1* KI mice. Extracellular field recordings showed that short-term synaptic plasticity, such as paired pulse ratio and post-tetanic potentiation was impaired in *Lsd1* KI mice, whereas long-term synaptic plasticity, including long-term potentiation and long-term depression, was normal. Moreover, the frequency of miniature excitatory postsynaptic current was significantly increased, suggesting presynaptic dysfunction in *Lsd1* KI mice. Consistent with this, RNA-seq analysis using the hippocampus of *Lsd1* KI mice showed significant alterations in the expressions of presynaptic function-related genes. Intriguingly, LSD1n-SA mutant showed diminished binding to histone deacetylase 1 (HDAC1) compared to LSD1n-WT in SH-SY5Y cells. These results suggest that LSD1 is involved in the regulation of presynaptic gene expression and subsequently regulates the hippocampus-dependent learning and memory in phosphorylation-dependent manner.

Disclosures: C. Lim: None. **H. Nam:** None. **J. Lee:** None. **D. Kim:** None. **J. Choi:** None. **S.J. Kang:** None. **S. Kim:** None. **H. Kim:** None. **C. Kwak:** None. **K. Shim:** None. **S. Kim:** None. **H. Ko:** None. **R. Lee:** None. **E. Jang:** None. **J. Yoo:** None. **J. Shim:** None. **M. Islam:** None. **Y. Lee:** None. **J. Lee:** None. **S. Baek:** None. **B. Kaang:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.05/UU43

Topic: H.01. Animal Cognition and Behavior

Support: NIH/NINDS 1RO1 NS088053-01A1NIH Grant EY

Title: The role of the ventral hippocampus in contextual fear conditioning

Authors: *J. A. GRAHAM[1], B. J. WILTGEN[2]
[1]Neurosci., [2]Ctr. for Neurosci., UC Davis, Davis, CA

Abstract: The hippocampus(HPC) is critical for the storage and retrieval of episodic and spatial memories. It is also necessary for the retrieval of contextual fear memories wherein a previously neutral place becomes associated with danger. However, since the HPC is not the site of storage for the context-shock association, it must communicate with other structures in order to drive memory retrieval. Current evidence strongly implicates the amygdala (AMY) as the site of storage for the CS-US association in fear memories. In the case of contextual fear conditioning, it is thought that the HPC provides the contextual information needed by the AMY to form the CS-US association and support memory retrieval. Projections to the AMY originate within the ventral 2/3 of the HPC and terminate in anterior portions of the BA (basal amygdala). Several studies suggest that this projection is necessary for the retrieval of contextual fear memories. Here, we confirm that VHC-BA neurons are restricted to a particular lamina within the VHC, and that optogenetic manipulation of VHC neurons can disrupt fear memory retrieval. We plan to extend these findings by first quantifying activity during context fear training and retrieval within the VHC-BA projection using a combination of c-fos imaging and the expression of retrograde tracers. We will then utilize the fos-tTA inducible tagging system to identify ensembles activated by contextual fear memory retrieval in VHC-BA projecting neurons and restrict optogenetic manipulations to these same cells. In doing so, we will determine whether context-specific activity in this projection is necessary and/or sufficient to drive fear memory retrieval.

Disclosures: J.A. Graham: None. **B.J. Wiltgen:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.06/UU44

Topic: H.01. Animal Cognition and Behavior

Support: RO1 NS088053 to B.J.W.

Title: Context fear learning triggers metaplasticity in the hippocampus

Authors: *J. N. KRUEGER[1], A. P. CRESTANI[2], B. J. WILTGEN[1,3]
[1]Ctr. for Neurosci., Univ. of California Davis, Davis, CA; [2]Neurosci. Grad. Program, Federal Univ. of Rio Grande do Sul, Rio Grande do Sul, Brazil; [3]Dept. of Psychology, Univ. of California, Davis, Davis, CA

Abstract: It is widely believed that NMDARs are required for learning and memory. However, several studies have shown that prior experience can reduce or eliminate the need for these receptors during subsequent learning. For example, animals trained on a hippocampus-dependent fear conditioning task are able to encode new fear memories in the absence of NMDAR activation. Results like these suggest that prior behavioral training modifies cellular plasticity mechanisms such that NMDARs are no longer required for learning. Current models of memory cannot explain this finding and the mechanisms that give rise to NMDAR-independent learning have yet to be identified. Therefore, the goal of the current experiment is to identify hippocampal plasticity mechanisms that are altered by behavioral experience and determine their contribution to NMDAR-independent learning.
Electrophysiological studies have identified several types of long-term potentiation (LTP) in the hippocampus that do not require NMDARs. These forms of plasticity can be mediated by metabatropic glutamate receptors (mGluR), kainite receptors (KR), or voltage-gated calcium channels (VGCC). The current experiments determined which of these mechanism mediates NMDAR-independent learning. To do this, mice underwent fear conditioning in context A followed by context B. We have shown that the former requires NMDAR while the later does not. During learning in context B, we infused antagonists for mGluRs, KRs or VGCCs into the hippocampus and examined the effects on memory. Our preliminary data indicate that memory for context B is only impaired when mGluR receptors are blocked by the antagonist MCPG prior to training in context B. Antagonists for NMDARs, KRs or VGCCs were ineffective. In addition, MCPG had no effect on learning in context A. Together these data suggest that learning about novel events requires NMDARs while learning about similar events requires mGluRs. Similar results have been found in the barrel cortex after whisker stimulation (Clem & Barth, 2008).

Disclosures: J.N. Krueger: None. **A.P. Crestani:** None. **B.J. Wiltgen:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.07/UU45

Topic: H.01. Animal Cognition and Behavior

Support: The German Research Foundation (DFG) project YO177/4-1

Title: Molecular mechanisms of working memory and possible involvement of TRPC channels

Authors: *A. REBOREDA[1,2], M. J. VALERO-ARACAMA[3,4], F. M. THEISSEN[2], A. ARBOIT[2], A. CORBU[3], M. YOSHIDA[1,2]
[1]Functional Architecture of Memory, Leibniz Inst. For Neurobio., Magdeburg, Germany; [2]Cognitive Neurophysiol., German Ctr. for Neurodegenerative Dis. (DZNE), Magdeburg,

Germany; [3]Ruhr Univ. Bochum, Bochum, Germany; [4]Inst. for physiology and pathophysiology, University of Erlangen, Germany

Abstract: Working memory (WM) tasks and temporal association tasks require the subject to hold information for a short period of time. This retention of information is thought to be done by persistent firing which is shown in vivo from areas active during these tasks (i.e.: hippocampal area, prefrontal cortex) during the retention period. However, it still remains unclear what mechanism supports persistent firing in vivo.
In contrast to the classical idea that recurrent synaptic network supports persistent firing, we and others have shown that TRPC channels can support persistent firing in individual neurons in vitro. TRPC channels are modulated by complex intracellular molecular pathways downstream of metabotropic G-protein coupled receptors. However, it was never systematically studied whether these modulations of TRPC channels are in agreement with the modulation of WM. To assess the plausibility of the idea that TRPC channels support WM, in the present work, we review molecular pathways supporting WM and persistent activity in vivo, and then we compare them to the mechanisms supporting persistent activity and TRPC channels in vitro.
We conclude that muscarinic (mainly M1), serotonin $5HT_{2A}$ and moderate activation of noradrenergic α_1 receptors, which are coupled to Gq and lead to PLC pathway activation, show a positive effect on WM and the maintenance of the persistent firing in vivo. While in vitro, the same pathway activates TRPC channels and trigger persistent firing in the areas engaged in WM activities. In addition, receptors coupled to Gi (M2, α_2, β_2, D_2) improve also WM and increase persistent firing in vivo, while in vitro experiments show that the Gi-triggered decrease of cAMP levels can activate TRPC channels and strengthen persistent firing. On the other hand, receptors coupled to Gs (β_1, $5HT_6$, D_1) impair WM and persistent firing in vivo, while the increase of cAMP levels leads to TRPC inhibition and suppression of the persistent firing observed in vitro. After reviewing and comparing the metabotropic pathways involved in WM and those leading to TRPC activation, we conclude that they show a high degree of homology and that makes TRPC channels strong candidates to be activated during the WM and temporal association tasks generating the persistent firing observed in vivo.

Disclosures: A. Reboreda: None. **M.J. Valero-Aracama:** None. **F.M. Theissen:** None. **A. Arboit:** None. **A. Corbu:** None. **M. Yoshida:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.08/UU46

Topic: H.01. Animal Cognition and Behavior

Support: The German Research Foundation (DFG) project YO177/4-1

Title: Investigating the role of TRPC channels in hippocampal persistent firing

Authors: *A. ARBOIT[1], F. THEISSEN[1], J. SCHWEIHOFF[2], L. BIRNBAUMER[3,4], M. YOSHIDA[1,5]
[1]Cognitive Neurophysiol., German Ctr. For Neurodegenerative Diseases-DZNE, Magdeburg, Germany; [2]Ruhr-Universität Bochum, Bochum, Germany; [3]Natl. Inst. of Envrn. Hlth. Sci., Research Triangle Park, NC; [4]Inst. of Biomed. Res. (BIOMED UCA-CONICET), Buenos Aires, Argentina; [5]Leibniz Inst. For Neurobio., Magdeburg, Germany

Abstract: The hippocampus plays an important role in memory tasks requiring an active maintenance of memory for a short period of time. Persistent firing, the ability of a neuron to maintain sustained firing after an input is terminated, is thought to support this phenomenon at the cellular level. In this study, we investigated the roles of transient receptor potential cation (TRPC) channels in supporting persistent activity in the hippocampal CA1 pyramidal cells focusing mainly on the TRPC4 and TRPC5 channels, which are highly expressed in the hippocampus. TRPC channels are one of the main candidates supporting persistent firing. However, their role in this phenomenon in the hippocampus is still not clear. Previous works in the prefrontal cortex using TRPC KO animals didn't show a clear link between persistent firing and these channels. Hence to give a conclusive answer, we choose to tackle the question combining different approaches, using pharmacological tools, antibodies and conditional KO of TRPC5. Our immunohistochemistry confirmed the expression of both TRPC4 and TRPC5 channels in the hippocampus. Using in vitro whole-cell patch clamp recordings in mouse hippocampal slices, we found that bath application of Carbachol (CCh), a cholinergic receptor agonist, induced persistent firing (PF) in about 70% of cells recorded. Frequency of persistent firing was around 9Hz and persistent firing typically lasted for 20 seconds. Then we investigated the role of TRPC channels. We started with a pharmacological approach using two novel TRPC channels blockers: clemizole hydrochloride and ML204. These drugs at high concentrations target both TRPC4 and TRPC5 while at lower concentrations ML204 targets mainly TRPC4 and clemizole hydrochloride targets TRPC5. Both of these blockers affected persisting firing, blocking it or reducing its length and/or frequency. In the second step, we used TRPC antibodies diluted in the intracellular solution to investigate how selectively blocking those channels could affect persistent activity. TRPC4 or TRPC5 antibodies suppressed PF in a similar way to the TRPC blockers. In the last approach we used a conditional TRPC5 KO mouse. Persistent firing in KO mice was greatly reduced compared to wild type mice leaving only a short depolarization. This depolarization plateau was almost completely suppressed by an application of ML204 or relatively high concentration of clemizole hydrochloride, suggesting an involvement of TRPC4. These results suggest that TRPC4 and TRPC5 channels may play a central role in supporting persistent firing in CA1 hippocampal cells.

Disclosures: A. Arboit: None. **F. Theissen:** None. **J. Schweihoff:** None. **L. Birnbaumer:** None. **M. Yoshida:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.09/UU47

Topic: H.01. Animal Cognition and Behavior

Support: The German Research Foundation (DFG) project YO177/4-1

Title: TRPC5 channels in the hippocampus support trace fear conditioning

Authors: *F. M. THEISSEN[1], A. ARBOIT[2], L. BIRNBAUMER[3,4], M. SAUVAGE[5], M. YOSHIDA[2]
[1]Cognitive Neurophysiol., [2]DZNE, Magdeburg, Germany; [3]Natl. Inst. of Envrn. Hlth. Sci., Research Triangle Park, NC; [4]Inst. of Biomed. Res. (BIOMED UCA-CONICET), Buenos Aires, Argentina; [5]Leibniz Inst. for Neurosci., Magdeburg, Germany

Abstract: Increasing evidence suggests the involvement of the hippocampus in temporal association tasks and working memory both in human and animals. Persistent firing has long been believed to be the cellular correlate of these tasks to support short-term (up to tens of seconds) retention of memory. However, the molecular mechanisms supporting persistent firing have not been identified. We have recently shown that hippocampal principal neurons support persistent firing driven by cholinergic receptors, and downstream activation of TRPC-like channels in vitro. To investigate the role of TRPC channels in temporal association memory, we tested a conditional gene-knockout (KO) confined to the hippocampus in male TRPC5 floxed mice, in a trace fear conditioning task. Conditional KO was induced by an injection of a viral vector containing Cre recombinase and a GFP reporter. A control group of animals was injected with a viral vector containing only a GFP reporter. Four weeks later, we first conducted in vitro recordings. We found that persistent firing in CA1 pyramidal cells was significantly reduced, indicating the supportive role of TRPC5 in persistent firing. Conditioning of the remaining animals was conducted as follows: After a baseline period, mice were presented with several tone-shock (CS-US, trace period of 30 seconds) pairings on day 1 in Context A, and were probed 24 hours later in Context B, and a further 24 hours later in context A. The two groups differed in their freezing levels throughout the conditioning session on Day 1, with the TRPC5 KO mice freezing significantly less, indicating that they struggled to learn the condition of the task. On Day 2, the KO group also showed significantly less freezing in response to the tone, indicating that they had failed to form the association between the tone and the shock. On Day 3 there was no significant difference between groups, and both groups freezing levels remained very low, indicating that the initial conditioning had led to an association between the CS and the US, instead of an association between the US and the context. Hereby we have shown that TRPC5, a

channel which is fundamental to the generation of sustained activity patterns, contributes to trace fear conditioning performance.

Disclosures: F.M. Theissen: None. **A. Arboit:** None. **L. Birnbaumer:** None. **M. Sauvage:** None. **M. Yoshida:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.10/UU48

Topic: H.01. Animal Cognition and Behavior

Title: Deficiency of TRPC1 exacerbated the apoptosis and cognitive impairment induced by Aβ

Authors: *M. LI[1], J. WANG[2]
[1]Pathophysiology, Huazhong Univ. of Sci. and Technol., Hubei, China; [2]Pathophysiology, Huazhong Univ. of Sci. and Technol., Wuhan, China

Abstract: Extracellular precipitation of β-amyloid (Aβ) is a pathological characteristic of Alzheimer's disease (AD). Canonical transient receptor potential (TRPC) channels are widely expressed throughout the nervous system while their functions remain largely unclear. Here, we studied the effects of TRPC1 on cognitive deficit induced by Aβ. We found that TRPC1 was dispensable for cognition of mice in physiological conditions. However, mice lacking of TRPC1 had a poorer performance in learning and memory when injected with Aβ in the unilateral ventricle. Treated with Aβ, TRPC1 knockout mice showed an increased level of apoptosis and a significant lossing of neurons in hippocampus. while deficiency of TRPC1 in physiological conditions did not affect the hippocampal apoptosis of mice. To further verify it, we overexpressed TRPC1 in cells and found it protected aganist to the apoptosis induced by Aβ treatment. On the other hand, we found that overexpression of TRPC1 reduced the production of Aβ in 293-APP cells, which overexpressed amyloid protein precursor stablly. These findings indicate that deletion of TRPC1 exacerbated the cognitive impairment induced by Aβ through apoptosis increasing.

Disclosures: M. Li: None. **J. Wang:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.11/UU49

Topic: H.01. Animal Cognition and Behavior

Support: National Basic Research Program of China (No. 2015CB553504)

Natural Science Foundation of China 81571302

Natural Science Foundation of China 81573404

Title: Methamphetamine activating hippocampal HCN channels involves in drug-associated context learning

Authors: *R. SONG[1], S.-Z. ZHANG[1], D.-N. CAO[1], L.-J. YANG[2], Z.-Y. WANG[1], N. WU[1], J. LI[1], J.-Q. ZHENG[1]
[1]Beijing Insistitute of Pharmacol. and Toxicology, Beijing, China; [2]Dept. of Anesthesia, Chinese PLA general hospital, Beijing, China

Abstract: Addictive drug-associated context learning plays an important role in the development of drug addiction, and changes in the electrophysiological properties and synaptic plasticity of hippocampal neurons are thought to be the cellular underpinnings of drug-context associative learning and memory. Hyperpolarization-activated, cyclic nucleotide-gated (HCN) channels play a fundamental role in controlling neuronal excitability and synaptic transmission in the hippocampus; however, whether HCN channels participate in context-associated learning remains unclear. In the present study, we found that methamphetamine (1 μM) increased the frequency of action potentials accompanied by a moderate depolarization of membrane potential in CA1 pyramidal neurons in hippocampal slices, which was abolished by blocking HCN channels, suggesting an involvement of HCN channels activation. Methamphetamine (0.1 and 1 μM) augmented the I_h amplitude by enhancing the conductance of HCN channels and shifting the activation curve to more depolarized potentials in hippocampal CA1 pyramidal neurons, thereby increasing the action potentials frequency. Furthermore, we found that the facilitation of HCN channels by methamphetamine was in a dopamine-dependent manner and probably also in a direct fashion. Functionally, the methamphetamine facilitation of HCN channels involved the long-term potentiation induction at Schaffer collateral-Cornu Ammonis1 excitatory synapses and the conditioned place preference acquisition, suggesting an important role of HCN channels in drug-associated context learning. In conclusion, these data supported that methamphetamine activated HCN channels in hippocampal pyramidal neurons and thus enhanced pyramidal membrane excitability and excitatory synaptic plasticity, which contributed to drug-associated context learning. These findings extend our understanding of the neurobiological mechanisms

underlying methamphetamine addiction. **Keywords:** Drug addiction, Hyperpolarization-activated cyclicnucleotide-gated channels, Methamphetamine, Drug-associated context learning, Hippocampal pyramidal neurons, Long-term potentiation

Disclosures: **R. Song:** None. **S. Zhang:** None. **D. Cao:** None. **L. Yang:** None. **Z. Wang:** None. **N. Wu:** None. **J. Li:** None. **J. Zheng:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.12/UU50

Topic: H.01. Animal Cognition and Behavior

Support: George E. Hewitt Foundation

DA025922

AG050787

AG051807

MH101491

Title: Role of CREST in synaptic plasticity and memory formation

Authors: *T. J. HEMSTEDT, E. A. KRAMÁR, Y. ALAGHBAND, D. P. MATHEOS, J. J. BANIHANI, M. A. WOOD
Neurobio. and Behavior, Univ. of California Irvine, Irvine, CA

Abstract: Epigenetic mechanisms have the potential to make persistent changes at the cellular level that can lead to long-term behavioral adaptations. Alterations of the epigenome have been shown to impact memory consolidation and to change gene expression profiles during memory consolidation. Chromatin remodeling is a major epigenetic mechanism, but it only recently gained attention in the learning and memory field. Chromatin remodeling complexes are multi-protein complexes that interact with DNA or chromatin structure and possess an ATP-dependent enzyme that induces a shift in the nucleosome positioning and ultimately regulate gene transcription. Recently, the Brg1/hBrm Associated Factor (BAF) complex has been gaining interest as it contains neuron-specific subunits and has been linked to intellectual disability disorders. Here, we investigate the neuron-specific subunit calcium-responsive transactivator (CREST) of the BAF complex. To knockdown the expression of CREST, we injected morpholinos and siRNA against CREST into the CA1 region of the dorsal hippocampus of mice.

Our results indicate that CREST is critical for synaptic plasticity and additionally a decrease in CREST expression leads to deficits in contextual fear memory and object-location memory.

Disclosures: T.J. Hemstedt: None. **E.A. Kramár:** None. **Y. Alaghband:** None. **D.P. Matheos:** None. **J.J. Banihani:** None. **M.A. Wood:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.13/UU51

Topic: H.01. Animal Cognition and Behavior

Support: NIH/NIDCD R03 DC014753-01

Start-up funding from the School of Arts and Science and Psychology Department at Rutgers University -- New Brunswick (KMB)

Title: HDAC3-inhibition facilitates long-term memory consolidation of sound-specific information

Authors: *A. SHANG, S. BYLIPUDI, K. M. BIESZCZAD
Behavioral and Systems Neuroscience, Dept. of Psychology, Rutgers Univ., Piscataway, NJ

Abstract: Neuroplasticity and successful long-term memory (LTM) can be modulated by epigenetic mechanisms, such as chromatin modification. Histone deacetylases (HDACs) typically oppose histone acetyltransferases (HATs) and repress gene expression by constricting chromatin. HDAC inhibitors have therefore been shown to open chromatin and facilitate various forms of LTM (e.g. McQuown & Wood, 2011; Malvaez et al., 2013). For example, rats treated with a pharmacological HDAC3-inhibitor (HDAC3i) during consolidation of an auditory associative task have enhanced memory for the behaviorally-salient sound-frequencies in a memory test after training, as well as expanded representation in primary auditory cortex (A1) for the signal-frequencies (determined by electrophysiological “mapping” of A1 tonotopy after all training and testing; Bieszczad et al., 2015). Here, we test the emerging hypothesis that HDAC3 controls the sensory-specificity of LTM (Phan & Bieszczad, 2016). Adult male rats (Sprague-Dawley) were trained on a two-tone frequency-discrimination (2TD) task with immediate post-training injections of an HDAC3i (RFGP966, Abcam Inc.; 10mg/kg, *s.c.*) or vehicle (Veh) for 3 consecutive days. Rats learned to associate one of two spectrally-different pure tones with reward over ~3 weeks (5.0 kHz, CS+; 11.5 kHz, CS-). HDA3i facilitated the rate of 2TD task acquisition: treated rats (N=12) reached performance criterion faster than vehicle controls (N=12). Furthermore, after both groups reached the *same level* of asymptotic performance, a stimulus generalization memory test (SGT) was administered. BP responses

elicited by different sound frequencies (including the CS+ & CS-) were recorded under extinction conditions. Groups differed in the proportion and latency of BPs to SGT test tones, which indicated that HDAC3i-treated rats had formed more *specific* memory for both CS frequencies compared to Veh-treated rats (N=9). A subset of animals also underwent a 2-week delayed SGT (HDAC3i, N=4; Veh, N=4) and were immediately re-tested for 2TD performance. Resilient performance in HDAC3i-treated rats weeks after HDAC3i injections suggest that limited treatment during initial task acquisition has lasting effects on frequency-specific memory. A1 recordings after immediate and 2-week delayed SGTs reveal how tonotopic representation of different associative sound-signals (CS+ and CS- frequencies) interacts in animals that have specific vs. non-specific memories. These data support that HDAC3 is key negative regulator of the sensory specificity of LTM formed.

Disclosures: **A. Shang:** None. **S. Bylipudi:** None. **K.M. Bieszczad:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.14/UU52

Topic: H.01. Animal Cognition and Behavior

Support: RSF grant №14-25-00072

RFBR grant №16-34-00918

Title: Histone acetylation controls transcription of the atypical protein kinases in primary neuronal cultures

Authors: *A. BORODINOVA, M. VOLOBUEVA, A. BOLSHAKOV, P. BALABAN
Inst. of Higher Nervous Activity, Moscow, Russian Federation

Abstract: Epigenetic modifications play a crucial role in learning and memory consolidation and storage. Some indirect evidences support the idea that atypical protein kinases (aPKCs), involved in memory consolidation and maintenance, may be the targets of epigenetic modifications. In the current study we are testing whether increased acetylation of histones, which usually occurs during learning, may determine transcriptional activity of aPKCs. Using real-time PCR method we have analyzed the dynamics of aPKCs expression in primary cortical neuron cultures incubated with histone deacetylase inhibitor trichostatin A (TSA). Histone acetylation affects the chromatin structure and usually enhances gene transcription. As it was expected, in the presence of TSA the mRNA levels of protein kinase C iota/lambda (PKCi/l) and protein kinase C zeta (PKCz) has temporarily increased. Surprisingly, the pattern of expression of protein kinase M zeta (PKMz) was characterized by gradual decrease with a minimum at 18-19 hours of

incubation with TSA. Moreover, similar changes in aPKCs expression were observed in cortical cultures, incubated with alternative histone deacetylase inhibitor sodium butyrate for 18-19 hours. It perfectly corresponds to our previous results and excludes the possibility of nonspecific activity of TSA. Application of the transcription inhibitor actinomycin D one hour prior to TSA treatment abolished the TSA-induced rise of PKCi/l and PKCz mRNA levels. Observed changes in PKMz expression during incubation with actinomycin require further investigation. Summarizing, our results suggest that histone acetylation status may directly determine the level of expression of atypical protein kinases in the neurons.

Disclosures: A. Borodinova: None. **M. Volobueva:** None. **A. Bolshakov:** None. **P. Balaban:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.15/UU53

Topic: H.01. Animal Cognition and Behavior

Title: Changes in nuclear geometry are associated with long-term memory formation

Authors: *I. CERA, A. ABENTUNG, C. REDDY, P. FEURLE, G. APOSTOLOVA, G. DECHANT
GE Neurowissenschaften, Innsbruck, Austria

Abstract: Special AT-rich sequence binding protein 2 (*SATB2*) is a risk locus for schizophrenia and encodes a highly conserved DNA-binding protein that regulates higher order chromatin configuration. We recently showed that deletion of Satb2 from adult mouse forebrain impairs long-term memory formation (Jaitner et al., 2016). At molecular level, we found that Satb2 interacts with proteins of the inner nuclear membrane including Lem2, Lem3, Baf and Lap2. Complexes of these lamina-associated proteins are known regulators of both gene transcription and nuclear shape. Therefore we have investigated the role of Satb2 in neuronal activity-dependent plasticity of nuclear morphology of hippocampal neurons. In hippocampal neuronal cultures synaptic activity stimulates the formation of nuclear infoldings (Wittman et al., 2009) and causes an up-regulation of Satb2 protein levels. We found that genetic deletion of Satb2 prevents these changes of nuclear morphology. Overexpression of Satb2 in vitro increased the number of nuclear infoldings in unstimulated cultures. To test whether the interaction between Satb2 and inner nuclear membrane proteins is required for nuclear infoldings, we used an RNAi approach to reduce Lem2 protein level. The experiment confirmed that the interaction between Satb2 and the nuclear envelope trans-membrane protein Lem2 is important for Satb2-induced changes in neuronal nuclei geometry. In vivo the number of nuclear infoldings is greatly reduced in the hippocampal CA1 field of $Satb2^{CamKIICre}$ conditional knock-out mice. Nuclear

dysmorphology is rescued together with long-term memory deficit when *Satb2* is re-expressed in the hippocampus of Satb2 conditional mutants. Based on our results we hypothesize that Satb2 in the neuronal nucleus modulates association of specific chromatin loops with the inner nuclear membrane and consequently regulates genes expression underlying the consolidation of memory.

Disclosures: I. Cera: None. **A. Abentung:** None. **C. Reddy:** None. **P. Feurle:** None. **G. Apostolova:** None. **G. Dechant:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.16/UU54

Topic: H.01. Animal Cognition and Behavior

Support: NS28912

MH73136

MH096889

Title: Multiple acute concurrent stresses and memory: Females are not protected

Authors: *A. K. SHORT[1], Y. CHEN[2], J. MOLET[4], J. C. LAUTERBORN[3], C. M. GALL[5], T. Z. BARAM[1]
[2]Pediatrics, [3]Dept Anat. & Neurobio., [1]Univ. of California Irvine, Irvine, CA; [4]CLINATEC, CEA MINATEC CAMPUS, Grenoble, France; [5]Anat. and Neurobio., Univ. of California, Irvine, CA

Abstract: Rationale: Multiple, concurrent acute stresses involving emotional, physical and social components enduringly impair spatial memory in male mice, which is associated with functional loss of synaptic potentiation and structural disruption of synapse-bearing dendritic spines in the hippocampus. Male and females behave differently in response to emotional and social stressors, and sex differences in spatial memory have long been observed in humans and animals. However, little is known about the underlying synaptic and molecular mechanisms responsible for these differences. Here, we subjected male and female mice to simultaneous multiple stresses lasting 2 h (MAS) and investigated the sex effects of MAS on memory function and their underlying synaptic basis. Methods: Male and female C57 and YFP transgenic mice were subjected to MAS separately and trained/tested in a hippocampus-dependent novel location object task 6 h, 24 h, and 1 week after the stress. Hippocampal slices from stressed mice and controls were used to test the different effects of MAS on the dendritic spines (YFP and Golgi staining on slices) and synapses represented by PSD-95 in field CA1, the location has been implicated to play a key role in place field and memory consolidation. Further experiments are in progress to identify the molecular basis responsible for the differences observed. Results:

Simultaneous acute stresses provoked profound, enduring memory problems in male mice. These resulted from lost ability of hippocampal synapses to sustain a potentiated state. In females, synapse loss was as profound as in males. Memory tests are in progress at the time of this submission.Conclusions: Modern life is rife with concurrent acute stresses. The consequences of these multiple acute stress clearly differ from those of a single acute stress in males. The current studies increase our understanding of sex-specific differences in the effects of multiple, acute, concurrent modern-life like stress on memory.

Disclosures: A.K. Short: None. **Y. Chen:** None. **J. Molet:** None. **J.C. Lauterborn:** None. **C.M. Gall:** None. **T.Z. Baram:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.17/UU55

Topic: H.01. Animal Cognition and Behavior

Support: NSFC Grant 81471101

Title: IQGAP1-ERK signaling regulates histone posttranslational modifications in fear memory

Authors: *C. GAO, X.-Y. LIU, B. YAO, L. JIN, N. SUN, J.-R. HAO
Sch. of Anesthesiol., Xuzhou Med. Univ., Jiangsu, China

Abstract: Epigenetic mechanisms in learning and memory are in particularly interesting in neuroscience recently. We previously found IQGAP1, a scaffolding protein of MAPK, was involved in fear memory via interacting with GluN2A subunits of NMDA receptors and the ERK cascade. But it's unknown whether histone posttranslational modifications is regulated by IQGAP1/ERK signal pathway. To shed light on specific functions of IQGAP1 in memory progresses and the precise mechanisms of this regulation, we performed in vivo studies using *IQGAP1*$^{-/-}$ and *IQGAP1*$^{+/+}$ mice. We found that *IQGAP1*$^{-/-}$ mice exhibited impaired fear memory as previously described. In addition, *IQGAP1*$^{-/-}$ mice showed decrease in ERK1/2 and histone H3S10 phosphorylation, H3K14 acetylation, as well as decreased levels of c-Fos and BDNF compared to their wild-type littermates after fear training. Correspondingly, disruption of the epigenetic regulation induced by ERK signaling with intrahippocampal injection of MEK antagonist U0126 or GluN2A-selective pharmacological antagonist NVP-AAM077 blockaded contextual-dependent memory, while no changes were noted with NR2B-selective antagonist Ro25-6981. SAHA, a HDACs non-specific inhibitor, or knock-down HDAC2 with shHDAC2-AAV in dorsal hippocampus could significantly rescued the impaired fear memory, H3S10 phosphorylation, H3K14 acetylation, as well as decreased levels of c-Fos and BDNF. Thus, we

believe that the GluN2A/IQGAP1/ERK-dependent mechanisms in histone posttranslational modifications could underlie multiple phases of memory processes.

Disclosures: C. Gao: None. **X. Liu:** None. **B. Yao:** None. **L. Jin:** None. **N. Sun:** None. **J. Hao:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.18/UU56

Topic: H.01. Animal Cognition and Behavior

Support: ADA Grant ADA1-17-PDF-138

NIH Grant NIH/DK093587

NIH Grant NIH/DK101379

USDA Grant USDA/Cris6250-51000-059-04S

NIH Grant NIH/DK099443

Title: Nuclear receptor corepressors regulate cognitive function by modulating GABA signaling

Authors: *W. ZHOU[1], Y. HE[2], S. HONG[1], G. DING[6], H. K. YALAMANCHILI[7], Y.-W. WAN[8], B. PAUL[3], C. WANG[1], Y. GONG[1], Q. WU[2], Q. TONG[9], Z. LIU[4], Y. XU[2], Z. SUN[5]
[1]Dept. of Medicine-Endocrinology, [2]USDA/ARS Children's Nutr. Res. Center, Dept. of Pediatrics, [3]Dept. of Mol. and Cell. Biol., [4]Dept. of Pediatrics, Jan and Dan Duncan Neurolog. Res. Institute, Texas Children’s Hospi, [5]Dept. of Medicine-Endocrinology, Dept. of Mol. and Cell. Biol., Baylor Col. of Med., Houston, TX; [6]The Intl. Peace Maternity & Child Hlth. Hosp. of China welfare institute, Shanghai, China; [7]Dept. of Pediatrics, Jan and Dan Duncan Neurolog. Res. Institute, Texas Children’s Hosp., Houston, TX; [8]Dept. of Mol. and Human Genetics, Dept. of Pediatrics, Baylor Col. of Medicine,Jan and Dan Duncan Neurolog. Res. Institute, Texas Children’s Hosp., Houston, TX; [9]Ctr. for Metabolic and Degenerative Disease, Inst. of Mol. Medicine, McGovern Med. Sch., Univ. of Texas Hlth. Sci. Ctr. at Houston, Houston, TX

Abstract: Nuclear Receptor Corepressors (NCORs) directly participate in the epigenomic action of various hormones and metabolic intermediates. NCORs are implicated in the pathogenesis of autism, but their role in cognitive function remains unclear. NCOR1 and NCOR2 (also known as SMRT) regulate gene expression by recruiting and activating histone deacetylase 3 (HDAC3) through the Deacetylase Activation Domain (DAD). Here we find that abolishing HDAC3 enzymatic activity with knock-in mutations in DAD (NS-DADm) led to mood alteration, social

avoidance, and cognitive dysfunction in mice. Depletion of NCORs specifically in GABAergic neurons recapitulated the cognitive dysfunction and memory deficit. Electrophysiology, optogenetics, chemogenetics, circuit mapping, functional genomics, ribosome profiling, and immunofluorescence analysis identified molecular and circuitry mechanisms through which NCORs regulate GABA signaling and hippocampal synaptic plasticity. These findings established NCORs as pivotal epigenomic regulators of GABA signaling and cognitive function.

Disclosures: W. Zhou: None. **Y. He:** None. **S. Hong:** None. **G. Ding:** None. **H.K. Yalamanchili:** None. **Y. Wan:** None. **B. Paul:** None. **C. Wang:** None. **Y. Gong:** None. **Q. Wu:** None. **Q. Tong:** None. **Z. Liu:** None. **Y. Xu:** None. **Z. Sun:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.19/UU57

Topic: H.01. Animal Cognition and Behavior

Support: NIH Grant R01NS057128

NIMH Grant R15MH101703

Title: Tat-PTEN1 enhances hippocampal LTP and learning & memory through selective inhibition of calpain-2-mediated PTEN cleavage

Authors: *Y. LIU[1], Y. WANG[1], J. SUN[1], Y. LUO[1], X. BI[2], M. BAUDRY[1]
[1]Western Univ. of Hlth. Sci., Pomona, CA; [2]Western Univ. Hlth. Sci., Pomona, CA

Abstract: Calpain-1 and calpain-2 play opposite roles in learning and memory: Calpain-1 activity is required for certain types of synaptic plasticity and learning and memory, while calpain-2 activation limits the extent of plasticity and learning during the consolidation period. During the consolidation period, calpain-2 cleaves PTEN, which stimulates mTOR-dependent synthesis of the ERK inhibitor, SCOP, thereby limiting the duration of ERK activation. A selective calpain-2 inhibitor (Z-Leu-Abu-CONH-CH2-C6H3(3,5-(OMe)2), enhances learning & memory by preventing calpain-2 mediated PTEN cleavage and prolonging ERK activation in dorsal hippocampus in mice. To further evaluate the role of calpain-2-mediated PTEN cleavage in learning and memory, we first determined the calpain-2 cleavage sites in PTEN. Two PTEN fragments generated by calpain-2 cleavage were analyzed by LC/MS/MS, which resulted in the identification of S364-V365 as one of the putative calpain-2 cleavage sites in PTEN. Five peptides based on PTEN sequence surrounding S364-V365 were synthesized. While none of the peptides inhibited overall calpain-1 or calpain-2 activity, one of them was directly cleaved by calpain-2 and they all partially blocked calpain-2-mediated PTEN cleavage. We selected

SSTSVTPDVSDN and linked it to the tat peptide to make it membrane-permeable, and this construct is referred to as tat-PTEN1. Tat-PTEN1 (10 μM) enhanced theta burst stimulation (TBS)-induced LTP, when applied for 90 min before TBS in hippocampal slices from WT mice. The effects of the tat-PTEN1 on fear conditioning learning and SCOP levels in dorsal hippocampus will be evaluated to confirm the role of calpain-2-mediated PTEN cleavage in L&M. These results further support the notion that calpain-2-mediated truncation of PTEN plays a critical role in the regulation of LTP and learning and memory in hippocampus. They further suggest that tat-PTEN1 could be further develop to enhance learning and memory under conditions associated with learning impairment.

Disclosures: Y. Liu: None. **Y. Wang:** None. **J. Sun:** None. **Y. Luo:** None. **X. Bi:** None. **M. Baudry:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.20/UU58

Topic: H.01. Animal Cognition and Behavior

Title: Baseline mTOR pathway phosphorylation is different within laser capture microdissected subregions of the medial temporal lobe in non-human primates

Authors: *B. A. CORGIAT[1,2], C. MUELLER[2], J. N. TURCHI[1], J. L. OLDS[3], R. C. SAUNDERS[1], L. A. LIOTTA[2], M. MISHKIN[1]
[1]Natl. Inst. of Mental Hlth., Bethesda, MD; [2]Ctr. for Applied Proteomics and Mol. Med., George Mason Univ., Manassas, VA; [3]George Mason Univ., Fairfax, VA

Abstract: The role of perirhinal cortex in visual recognition memory is well established, as is the role of cholinergic activation of perirhinal M1 muscarinic receptors. Neurophysiological data demonstrate that successful visual memory formation is characterized by enhanced multiunit activity in the upper-middle and deep layers of perirhinal cortex. However, the M1 muscarinic dependent intracellular signaling profiles underlying the critical synaptic changes that occur during long term visual memory formation are unclear. Long term memory formation depends upon protein translation and synaptic remodeling, both of which are regulated by the mTOR pathway. Here, we explore the mTOR signaling pathway in laser capture microdissected subregions of medial temporal lobe structures in normal monkeys.
Snap frozen samples of area TE, perirhinal (PRh), entorhinal (ERh), and hippocampal tissue from four Rhesus monkeys were cryosectioned and stained for Nissl substance. Laminar lysates were prepared by laser capture microdissecting specific layers of interest: layers III and V/ VI from PRh and area TE; layers II/III and V/VI from ERh; the stratum pyramidale (*str. pyr.*), the stratum radiatum (*str. rad.*) plus the stratum lacunosum-moleculare (*str. l-m.*) from regions CA1

and CA3, and the stratum granulosum (*str. gr.*) and stratum moleculare (*str. mol.*) of the dentate gyrus. In addition, whole region lysates spanning all cortical layers were prepared. The laminar and whole region lysates were printed onto protein microarrays, which enable a quantitative and multiplexed analysis of protein levels and phosphorylation states. Averaged over four monkeys, ERh displayed the highest mTOR pathway activation in whole region lysates, followed by PRh, area TE and the hippocampus. In laminar lysates, differential mTOR pathway activation patterns were revealed in the various cortical regions. For example, ERh layers II/III exhibit significantly increased phosphorylation levels along the mTOR pathway [e.g. mTOR S2481, p70S6K T412, rpS6 S235/S236, and eIF4E S209] compared to ERh layers V/VI. In contrast, PRh layers V/VI saw increased phosphorylation levels in eIF4E S209 and eIF4G S1108 compared to PRh layer III. In the hippocampus, increased levels of p70S6K S371, rpS6 S235/236, 4EBP1 T37/T46, and eIF4G S1108 were seen in the *str. pyr.* of CA1 and CA3 compared to their respective levels in *str. rad*. and *str. l-m.*; the dentate gyrus trended similarly with increased phosphorylation in *str. gr.* compared to *str. mol.* Using laser capture microdissection, we were able to demonstrate significant differences in mTOR signaling pathway activation in cortical layers implicated in memory formation.

Disclosures: **B.A. Corgiat:** None. **C. Mueller:** None. **J.N. Turchi:** None. **J.L. Olds:** None. **R.C. Saunders:** None. **L.A. Liotta:** None. **M. Mishkin:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.21/UU59

Topic: H.01. Animal Cognition and Behavior

Support: CIHR MOP-10848

CRC 950-201144

FRQS 32064

Title: mTORC1-mediated late LTP in somatostatin interneurons regulates hippocampal network plasticity and memory precision

Authors: *J. ARTINIAN[1,2], A. JORDAN[1,2], A. KHLAIFIA[1,2], A. LA FONTAINE[1,2], I. LAPLANTE[1,2], J.-C. LACAILLE[1,2]
[1]Dept of Neuroscience, Univ. of Montreal, Montreal, QC, Canada; [2]GRSNC, Montreal, QC, Canada

Abstract: Long-term synaptic plasticity is a prime candidate cellular substrate for learning and memory which remains largely unexplored in inhibitory interneurons. In the hippocampal CA1

region, excitatory synapses onto somatostatin interneurons (SOM-INs) show cell type-specific long-term potentiation (LTP) that regulates hippocampal network plasticity, can persist 24h and requires translation via Mechanistic Target Of Rapamycin Complex 1 (mTORC1). The present study aimed at investigating the functional role of translation-dependent LTP in SOM-INs in hippocampal networks and memory, by knocking out the expression of Raptor, an essential component of mTORC1, selectively in SOM-INs (SOM-Raptor-KO mice).
We first determined that treatment with mGluR1a agonist failed to increase ribosomal S6 protein phosphorylation, a downstream effector of mTORC1, and to induce persistent LTP in SOM-INs from SOM-Raptor-KO mice. Thus, SOM-INs show impairment in mTORC1 signalling and persistent synaptic plasticity in SOM-Raptor-KO mice. We next investigated the behavioral relevance of mTORC1-mediated persistent LTP. We showed with whole-cell recordings 24h after contextual fear conditioning (CFC) that training induces an increase in spontaneous and minimally evoked excitatory transmission, as well as input-output function at SOM-INs synapses in WT mice. SOM-INs dendritic spine density was also increased following training. These changes were prevented in SOM-INs from SOM-Raptor-KO mice, demonstrating that CFC induces persistent mTORC1-dependent LTP at SOM-INs synapses. At the network level, field recordings revealed upregulation of pyramidal cell Schaffer collateral pathway LTP by late LTP induction in SOM-INs which was prevented in SOM-Raptor-KO mice, indicating impairment in the regulation of CA1 network metaplasticity by SOM-INs LTP in SOM-Raptor-KO mice. At the behavioral level, SOM-Raptor-KO mice showed impaired long-term spatial reference and fear memory, and context generalization in CFC, but intact long-term cued-fear memory. These results indicate impairments in hippocampal-dependent memory in SOM-Raptor-KO mice.
Finally, we used transgenic mice with a knock-down of the upstream repressor of mTORC1, Tuberous Sclerosis Complex 1 (TSC1), selectively in SOM-INs (SOM-$TSC1^{+/-}$ mice) and found increased contextual fear memory but impaired context discrimination in these mice, indicating that increasing mTORC1 function in SOM-INs was sufficient to modulate hippocampal memory consolidation.
Our results suggest that learning-induced persistent mTORC1-LTP in SOM-INs regulates CA1 local network plasticity and hippocampal memory.

Disclosures: **J. Artinian:** None. **A. Jordan:** None. **A. Khlaifia:** None. **A. La Fontaine:** None. **I. Laplante:** None. **J. Lacaille:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.22/UU60

Topic: H.01. Animal Cognition and Behavior

Support: NIH R01NS085093

NIH R00 NS076364

NIH U54HD079124

NIGMS R25GM109442

BBSRC-BB/H018344/1

Title: The temporal dynamics of Arc expression regulate cognitive flexibility

Authors: *A. M. MABB[1], M. J. WALL[2], D. R. COLLINS[2], S. L. CHERY[1], Z. D. ALLEN[1], E. D. PASTUZYN[3], V. D. NIKOLOVA[4], S. S. MOY[5], B. D. PHILPOT[6], J. D. SHEPHERD[3], M. D. EHLERS[7], S. A. CORREA[8]
[1]Georgia State Univ., Atlanta, GA; [2]Univ. of Warwick, Coventry, United Kingdom; [3]Neurobio. and Anat., Univ. of Utah, Salt Lake City, UT; [4]Univ. of North Carolina, Chapel Hill, NC; [5]Psychiatry, Univ. of North Carolina at Chapel Hill Sch. of Med., Chapel Hill, NC; [6]Cell Biol. and Physiol., Univ. North Carolina, Chapel Hill, NC; [7]Res. and Develop., Biogen, Cambridge, MA; [8]Univ. of Bradford, Bradford, United Kingdom

Abstract: Neuronal activity regulates the transcription and translation of the immediate early gene Arc/Arg3.1, a key mediator of synaptic strength and plasticity. Ubiquitin-directed proteasomal degradation of Arc tightly limits its temporal expression, yet the significance of this regulation remains unknown. Here we have disrupted the temporal control of Arc removal by creating an Arc knock-in mouse (ArcKR) where the predominant Arc ubiquitination sites are mutated. In ArcKR neurons, induction of Arc with the group I mGluR selective agonist DHPG resulted in persistent accumulation of Arc protein. Consistent with the increased longevity of Arc protein, ArcKR mice have enhanced α-amino-3-hydroxy-5-methyl-4-isoxazolepriopionic acid (AMPA) receptor internalization and metabotropic glutamate receptor-dependent long-term depression (mGluR-LTD). Although ArcKR mice have no obvious motor or hippocampal-dependent spatial memory impairments, they exhibit deficits in selecting the strategy to perform a reversal task. This cognitive inflexibility is coupled to an increase in hippocampal Arc expression immediately following behavioral training and with a reduction in the threshold to induce mGluR-LTD. These findings suggest that the abnormal persistence of Arc protein limits the dynamic range of Arc-mediated AMPA receptor endocytosis during reversal learning, thereby reducing cognitive flexibility. Our work illuminates how the precise temporal control of activity-dependent molecules, such as Arc, regulate synaptic plasticity and is particularly crucial for cognitive flexibility.

Disclosures: **A.M. Mabb:** None. **M.J. Wall:** None. **D.R. Collins:** None. **S.L. Chery:** None. **Z.D. Allen:** None. **E.D. Pastuzyn:** None. **V.D. Nikolova:** None. **S.S. Moy:** None. **B.D. Philpot:** None. **J.D. Shepherd:** None. **M.D. Ehlers:** A. Employment/Salary (full or part-time):; Biogen. **S.A. Correa:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.23/UU61

Topic: H.01. Animal Cognition and Behavior

Title: Enhancement of synaptic plasticity by NYX-2925: The role of receptor trafficking and intracellular signaling

Authors: *R. A. KROES[1,2], M. E. SCHMIDT[1], S. U. SAHU[1], J. S. BURGDORF[1,2], M. A. KHAN[1], J. R. MOSKAL[1,2]
[1]Aptinyx, Inc., Evanston, IL; [2]Falk Ctr. for Mol. Therapeut., Northwestern Univ., Evanston, IL

Abstract: Aptinyx has developed a novel class of small molecule N-Methyl-D-Aspartate (NMDA) receptor modulators with broad applicability across neurologic and psychiatric disorders. It binds with high affinity to all four NR2 subtypes, rapidly enhances LTP, and improves learning and memory in rats. In addition, NYX-2925 (1 mg/kg, PO) induced significant alterations in structural plasticity, as measured by mature dendritic spine formation 24 hr post-dosing. To evaluate the rapid-acting effects of NYX-2925, an unbiased proteomic strategy was used to elucidate the biochemical processes that underlie its unique mechanism of action.
Rats were treated with either vehicle (0.5% CMC in saline; 1ml/kg) or NYX-2925 (1 mg/kg, PO). Medial prefrontal cortical proteins were isolated from animals sacrificed at 15, 30, 60 min, and 24 hr post-dosing. Both total and phosphopeptides enriched by HILIC fractionation were analyzed using nano-LC MS/MS analysis. The functional roles of the identified proteins were evaluated using Ingenuity Pathway Analyses.
The most robust proteomic changes in both phosphorylated and unmodified proteins, following administration of NYX-2925, were in signaling pathways associated with synaptic plasticity (LTP and LTD) and glutamatergic receptor trafficking (including clathrin-mediated endocytosis signaling, 14-3-3-mediated signaling, ephrin receptor signaling, protein ubiquitination, CDK5 signaling, aryl hydrocarbon receptor signaling, and semaphorin signaling).
To gain mechanistic insight into NYX-2925-mediated synaptic changes, proteomic analyses were performed on PSD95-coimmunoprecipitated proteins from drug-treated rats. Significant increases in cortical PSD95-associated GRIA2 and NMDAR2A at 60 min and 24 hours following drug administration were observed.
These results suggest that the cognitive-enhancing effects of NYX-2925 are driven in part by the modulation of trafficking and insertion of AMPARs and NMDARs into the synaptic PSD complex which, in turn, enhances LTP-associated synaptic plasticity.

Disclosures: R.A. Kroes: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx,

Inc. **M.E. Schmidt:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx, Inc. **S.U. Sahu:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx, Inc. **J.S. Burgdorf:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx, Inc. **M.A. Khan:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx, Inc. **J.R. Moskal:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx, Inc..

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.24/UU62

Topic: H.01. Animal Cognition and Behavior

Support: NINDS Grant R01NS057128 (MB)

NIMH Grant R15MH101703 (XB)

Daljit & Elaine Sarkaria Chair (XB)

LMU Faculty Research Grant (MF)

Title: Impaired cerebellar plasticity and eye-blink conditioning in calpain-1 knock-out mice

Authors: *M. R. FOY[1], S. HEYSIEATTALAB[2], K.-H. LEE[2], Y. LIU[2], X. BI[2], M. BAUDRY[2]
[1]Dept. of Psychology, Loyola Marymount Univ., Los Angeles, CA; [2]Western Univ. of Hlth. Sci., Pomona, CA

Abstract: Our recent studies indicate that calpain-1 activation is required for hippocampal synaptic plasticity, including long-term depression (LTD) and long-term potentiation (LTP) in field CA1, while calpain-2 activation limits the extent of potentiation during the consolidation period. However, little is known regarding the contributions of calpain-1 and calpain-2 to cerebellar synaptic plasticity, and in particular to LTP and various forms of LTD at synapses from the parallel fibers to the Purkinje cells. Low frequency (LFS, 1 Hz, 5 min)-induced LTP at these synapses was markedly impaired in cerebellar slices from calpain-1 knock-out (ko) mice. Application of a selective calpain-2 inhibitor improved LFS-induced LTP in both wild-type (wt) and calpain-1 ko mice, suggesting that, like in hippocampus, calpain-1 and calpain-2 play opposite functions in cerebellar LTP. Three different protocols were used to induce LTD at these synapses: LFS (1 Hz, 15 min), application of K-Glu (5 min) and application of the mGluR1 agonist, DHPG (10 min). All three forms of LTD were impaired in calpain-1 ko mice. DHPG

application was found to stimulate calpain-1 in cerebellar slices and DHPG-induced LTD impairment was reversed by application of a PP2A inhibitor, okadaic acid. To investigate the role of calpain-1 in a well-studied form of behavioral associative learning, we compared calpain-1 ko mice vs. wt mice in cerebellum-dependent delay eyeblink conditioning (EBC). Calpain-1 ko mice exhibited significant learning impairment in EBC during the first 2 days of acquisition training. However, after 5 days of training, the percentage of CRs between ko vs. wt mice was identical. During extinction, both calpain-1 ko and wt mice exhibited typical extinction patterns and there were no significant differences between these two groups. Our results indicate that calpain-1 plays critical roles in a variety of forms of synaptic plasticity and learning and memory in both the hippocampus and cerebellum, and that some of the signaling pathways underlying these forms of plasticity and learning are similar in both structures.

Disclosures: M.R. Foy: None. **S. Heysieattalab:** None. **K. Lee:** None. **Y. Liu:** None. **X. Bi:** None. **M. Baudry:** None.

Poster

428. Learning and Memory: Molecules and Mechanisms I

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 428.25/UU63

Topic: A.02. Postnatal Neurogenesis

Title: GABAergic modulation of cells within the neurogenic niche of the postnatal spinal cord

Authors: *N. SHAFIN[1,2], J. DEUCHARS[1], S. A. DEUCHARS[1]
[1]Sch. of Biomed. Sci., Univ. of Leeds, Leeds, United Kingdom; [2]Sch. of Med. Sci., Universiti Sains Malaysia, Health Campus Kubang Kerian, Malaysia

Abstract: Modulation of GABA mediated neuronal inhibition is one of the most important approaches for the treatment of CNS diseases. Precise targeting of such treatments depends on identification and characterisation of the different subunit complexes that exist. In the CNS, there are pools of neural stem cells (NSC), which can differentiate to become neurones, astrocytes or oligodendrocytes while progenitor cells have limited lineage. In addition to brain regions, where NSCs are now known to exist, the spinal cord area also has a neurogenic potential. Ependymal cells (ECs) of the brain and spinal cord respond to GABA. One approach to develop pharmacological treatments to modulate stem cell activity is to determine how endogenous proteins and receptors on NSCs can regulate their behaviour. Whole cell patch clamp recordings were made from cells in acute spinal cord slices. Responses to GABA were determined and compounds known to modulate GABA receptors in a subunit selective pattern were tested. Bath and puff application of GABA receptor agonists and antagonists were able to modulate the GABA receptor activation. FGIN-27; 1 μM (an agonist acting at a peripheral benzodiazepine receptor, called translocator protein (TSPO)) significantly enhanced responses to GABA in

ependymal cells. Octadecaneuropeptide (ODN), the endogenous ligand of the central benzodiazepine receptor alone caused both depolarising and hyperpolarising effects on ECs but did not significantly affect responses to GABA. Local ODN application caused fast hyperpolarisations that were mimicked by local application of low (2.5 μM) concentrations of GABA. To further characterise the GABA receptor, agonists and antagonists were used to enhance and block the receptor accordingly. Since TPMPA antagonised responses to ODN and low concentrations of GABA, this suggests that GABA receptors contain subunits from both GABAA and GABAC. Cells that responded to these modulators were recovered and viewed using fluorescence method and diaminobenzadine staining. The cells viewed were confirmed as ECs. If cells within the central canal area responded to GABA, it is possible that following an injury or onset of pathological condition, GABA and endogenous ligands such as ODN could be released in this area to modulate proliferation and differentiation. These experiments will help to confirm the role of GABA receptors in spinal cord cell proliferation.

Disclosures: **N. Shafin:** None. **J. Deuchars:** None. **S.A. Deuchars:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.01/UU64

Topic: H.01. Animal Cognition and Behavior

Support: SATT AxLR Maturation Grant

Title: Blockade of the sigma-1 receptor prevent brain plasticity induced in mice by habituation to a complex environment, the Hamlet test

Authors: *L. CROUZIER, T. MAURICE
INSERM UMR-S1198, Montpellier, France

Abstract: The σ_1 receptor is a membrane-associated protein expressed in neurons and glial cells. It is highly expressed at mitochondria-associated endoplasmic reticulum (ER) membranes (MAMs), complexing the glucose-related protein 78/binding immunoglobulin protein (BiP). Upon cellular stresss, or via agonist stimulation, σ_1 receptor dissociates from BiP and binds inositol 1,4,5-trisphosphate (IP_3) receptor, enhancing calcium entry into mitochondria. It can also translocate to plasma membrane and modulate the activity of numerous ionophores and receptors for neurotransmitters and trophic factors. The σ_1 receptor shapes cellular plasticity also by directly modulating the activity of pleiotropic transcription factors, such as NFKB, CREB or c-fos, involved in the modulation of pro- and anti-inflammatory genes, cell death and survival. The σ_1 receptor-mediated neuromodulation, affecting several cellular pathways has an important role on brain plasticity, response to stress, learning and memory, and neuroprotection. For instance,

its activation potentiates long-term potentiation, neurite outgrowth and hippocampal neurogenesis. We here analyzed its impact on brain plasticity induced by habituation to a complex enriched environment. The Hamlet test is a novel behavioral analysis appliance, fully automatized (ViewPoint, Lissieu, France), that provides a complex environment for testing topographical memory and spatio-temporal disorientation in mice. The apparatus mimics a small village with a central agora and streets expanding from it, leading to functionalized houses (Drink, Eat, Play, Run, Interact). Environmental enrichment is induced by habituating animals in the Hamlet, in groups of 6/8 individuals, during 4 h a day, for several weeks. Memory can be tested by depriving mice from water and testing their ability to locate the Drink house. Several groups were analyzed in parallel (non-habituated; habituated; habituated and repeatedly administered with NE-100, a selective σ_1 antagonist) and the expression and activity of the σ_1 receptor were analyzed after habituation. The antidepressant and anti-amnesic effect of selective σ_1 agonists, the hippocampal neurogenesis and trophic factor levels were analyzed. We report that the pharmacological inactivation of σ_1 receptor following NE-100 treatment prevented topographic memory, resistance to forced swimming and increased neurogenesis induced by habituation. The data showed that σ_1 receptor activity plays a major role in the consequences of enriched environment and therefore suggest that MAM physiology is impacted by enriched environment.

Disclosures: L. Crouzier: None. **T. Maurice:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.02/UU65

Topic: H.01. Animal Cognition and Behavior

Title: NYX-783: A novel small molecule NMDA receptor modulator with therapeutic potential for the treatment of post-traumatic stress disorder

Authors: *E. M. COLECHIO[1,4], T. BHATTACHARYA[4], J. S. BURGDORF[2], J. DUNNING[4], A. L. GROSS[6], J. M. PRIEBE[5], M. A. KHAN[4], P. K. STANTON[7], X.-L. ZHANG[7], C. CEARLEY[6], J. R. MOSKAL[3]
[1]Dept Neurosci, [2]Falk Ctr. for Mol. Therapeutics, McCormick Sch. of Engin., [3]Northwestern Univ., Evanston, IL; [5]Behavioral Pharmacol., [4]Aptinyx, Inc., Evanston, IL; [6]Aptinyx Inc, Evanston, IL; [7]Cell Biol. & Anat., New York Med. Col., Valhalla, NY

Abstract: Aptinyx has developed a novel class of small molecule N-Methyl-D-aspartic acid receptor (NMDAR) modulators with broad applicability across neurologic and psychiatric disorders. In the present study, NYX-783 was evaluated for efficacy in several animal models of learning and memory and of psychiatric disorders relevant to post-traumatic stress disorder

(PTSD). NYX-783 is an NMDAR modulator able to bind to all 4 NMDAR2 subtypes. NYX-783 was shown to enhance the magnitude of LTP at Schaffer collateral/CA1 synapses from hippocampal slices at 24 hours and 1 week post-dosing in vivo. NYX-783 was found to have high oral bioavailability and brain penetration, and showed no adverse effects in the Rotarod assay. NYX-783 showed an acute and long-lasting antidepressant-like effect in the forced swim assay: NYX-783 significantly decreased floating time vs. vehicle at all doses (1 ng/kg- 1 mg/kg) and this response was maintained up to 1 week post-dosing. To assess the therapeutic potential of NYX-783 for treating PTSD, NYX-783 was evaluated in a contextual fear conditioning assay. Rats were dosed with NYX-783 or vehicle 1 h before the first extinction trial and were assessed for freezing behavior on days 1-7 and 14. Treatment with NYX-783 decreased freezing relative to vehicle on days 2-4 and 14 demonstrating that NYX-783 is effective in facilitating fear extinction as well as enhancing consolidation of extinction learning. NYX-783 facilitated the extinction of contextual fear, suggesting that NYX-783 enhanced a synaptic plasticity-like mechanism associated with learning and memory and has therapeutic potential for the treatment of PTSD.

Disclosures: **E.M. Colechio:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **T. Bhattacharya:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **J.S. Burgdorf:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **J. Dunning:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **A.L. Gross:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **J.M. Priebe:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **M.A. Khan:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **P.K. Stanton:** F. Consulting Fees (e.g., advisory boards); Aptinyx Inc.. **X. Zhang:** None. **C. Cearley:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc. **J.R. Moskal:** A. Employment/Salary (full or part-time):; Aptinyx Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aptinyx Inc..

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.03/UU66

Topic: H.01. Animal Cognition and Behavior

Support: UNAM, DGAPA, PAPIIT IN307711

Title: Amygdala c-Fos expression in rats with tolerance to anxiolytic effect of midazolam

Authors: *D. J. GONZÁLEZ-SÁNCHEZ, SR, G. CASTILLO-ROBERTO, J. C. P. ARRIAGA-RAMÍREZ, S. E. CRUZ-MORALES
Psychopharmacology, UNAM, Tlalnepantla de Baz, Mexico

Abstract: Benzodiazepines (BZD) are prescribed mainly for treatment of anxiety and sleep disorders. Repeated administration of BZD induces the development of associative and pharmacological tolerance. C-Fos gene expression is a marker of neural activity, therefore could be a useful tool in the study of two different types of tolerance. The objective of present study was to assess the anxiolytic effect of midazolam (M), the development of associative and pharmacological tolerance, and the C-Fos expression in amygdala nuclei (CeA, LA, MePD and MePV) in rats administrated acutely or chronically in different contexts and placed on an Elevated Plus Maze (EPM).
Wistar male rats (200 g) were randomly assigned to five independent groups (N=8 rats in each group). A control group received saline; two groups were injected acutely with M in different contexts, colony (aMC) and laboratory (aML); two groups were injected for 20 days with M in different contexts, colony (cMC) and laboratory (cML). The groups received either acute or chronic administrations of 1 mg/kg, ip of M 15 min before the exposure to the EPM, the chronic groups were evaluated in the EPM on day 21 of treatment. The percentage of number of entries and time of permanence in open arms were considered an index of anxiety. Two h after EPM exposure, subjects were sacrificed and the c-Fos protocol was followed.
An anxiolytic effect was detected, and pharmacological and associative tolerance to M. C-Fos expression in CeA and LA was lower in acutely administrated groups and in cMC, conversely, C-Fos expression in cML was similar to that in the control group. There were not significant differences in C-Fos expression in MePD and MePV. Results suggest that CeA and LA are involved in the anxiety process.

Disclosures: D.J. González-Sánchez: None. **G. Castillo-Roberto:** None. **J.C.P. Arriaga-Ramírez:** None. **S.E. Cruz-Morales:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.04/UU67

Topic: H.01. Animal Cognition and Behavior

Support: NIH Grant R35 CA197289

Title: Chemotherapy-induced cognitive deficits in an APOE mouse model

Authors: *A. SPEIDELL[1], T. DEMBY[2], Y. LEE[3], O. RODRIGUEZ[4], C. ALBANESE[4], J. MANDELBLATT[5], G. REBECK[1]
[1]Neurosci., [2]Dept. of Tumor Biol., [4]Oncology, [5]Med., [3]Georgetown Univ. Med. Ctr., Washington, DC

Abstract: Cancer-related cognitive impairment is being increasingly reported among breast and other cancer survivors, although risks for this outcome are not fully understood. The apolipoprotein E4 (APOE4) allele, the strongest genetic risk factor for Alzheimer's disease, has been identified as one potential genetic risk factor for cancer-related cognitive impairment. Understanding risks and mechanisms related to diverse therapies is difficult in human populations. While there have been some important animal models of cancer-related cognitive impairment, none have examined gene-treatment interactions. We evaluated whether a battery of rodent behavioral assays could identify cognitive deficits in APOE knock-in C57BL/6 mice after treatment with chemotherapeutic drug. Twenty-four three-month old tumor-naïve mice (APOE3 n=11; APOE4 n=13) were subjected to a series of behavioral tests both before and after a single 10 mg/kg tail vein injection of Doxorubicin, a topoisomerase IV inhibitor used in treatment of breast cancer. Pre- and post-treatment open field and elevated plus tests demonstrated significantly decreased rearing behavior and willingness to explore open environments in both APOE3 and APOE4 mice following chemotherapy. Using the Barnes maze to measure post-treatment spatial learning and memory, there was significantly greater primary latency to the target hole (spatial learning) in the APOE4 mice compared to the APOE3 mice. Treated APOE4 mice showed similar impairment in recall of the target hole location in a 72-hour probe trial of the Barnes Maze (spatial memory). A marble burying assay and a novel object recognition test did not detect significant differences before and after Doxorubicin treatment in either group of APOE mice. These results suggest that chemotherapy has an adverse effect on cognitive behaviors, and that this may be an excellent model to elucidate how APOE genotype interacts with chemotherapy to affect behavior, brain structure, and brain biochemistry.

Disclosures: **A. Speidell:** A. Employment/Salary (full or part-time):; Employment, FT. **T. Demby:** None. **Y. Lee:** A. Employment/Salary (full or part-time):; Georgetown University. **O. Rodriguez:** A. Employment/Salary (full or part-time):; Employment, FT. **C. Albanese:** A.

Employment/Salary (full or part-time):; Employment, FT. **J. Mandelblatt:** A. Employment/Salary (full or part-time):; Employment, FT. **G. Rebeck:** A. Employment/Salary (full or part-time):; Employment, FT.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.05/UU68

Topic: H.01. Animal Cognition and Behavior

Support: ZR2014HL091 (Shandong, China, to FZ)

Foundation of Overseas Distinguished Taishan Scholars of Shandong Province (Shandong, China, to HZ)

Title: 6-Gingerol attenuates LPS-induced memory impairment via its anti-inflammatory and antioxidant activity

Authors: F. ZHANG[1], J.-G. ZHANG[1], X.-Q. HOU[1], Y.-M. ZHOU[1], *H. ZHANG[2,1]
[1]Institute of pharmacology, Taishan Med. Univ., Taian, China;[2]Depts Behav Med. & Psych, Pharm, West Virginia Univ. Hlth. Sci. Ctr., Morgantown, WV

Abstract: 6-Gingerol, a major component of gingerols extracted from *Zingiber officinale*, has been shown to exhibit anti-inflammatory and antioxidant bioactivities. Since neuroinflammation plays an important role in neurodegenerative diseases such as Alzheimer's disease, it was of interest to know if 6-gingerol improved memory via its anti-inflammatory and antioxidant properties. This was addressed using lipopolysaccharide (LPS)-induced changes in vitro in C6 cells and in vivo in rats. LPS at 100 µg/ml decreased cell viability, which was blocked by 6-gingerol (20 µM). Similarly, 6-gingerol reversed LPS (20 µg/ml)-induced increases in reactive oxygen species (ROS), nitrite (NO), inducible nitric oxide synthase (NOS2), tumor necrosis factor α (TNF-α), and interleukin-6 (IL-6) in C6 cells. Consistent with the in vitro effects, 6-gingerol (1 and 2 mg/kg) reversed LPS (2 µg, icv)-induced increases in latency to the platform during the acquisition training and decreases in the time spent in the target quadrant in the probe trial in the rat water-maze test, suggesting memory enhancement. These data suggest that 6-gingerol improves memory most likely via its anti-inflammatory and antioxidant effects and could be used for treatment of neurodegenerative disorders such as Alzheimer's disease.

Disclosures: F. Zhang: None. **J. Zhang:** None. **X. Hou:** None. **Y. Zhou:** None. **H. Zhang:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.06/UU69

Topic: H.01. Animal Cognition and Behavior

Support: DA033373

Title: Informative cues and drug reward modulate risky decision-making in a probabilistic foraging task

Authors: *A. P. SMITH[1], J. BECKMANN[2]
[1]Psychology, Univ. of Kentucky Dept. of Psychology, Lexington, KY; [2]Psychology, Univ. of Kentucky, Lexington, KY

Abstract: Many decisions involve uncertainty in regard to their potential outcome, and the ability of an individual to learn from such decisions requires flexible value updating from trial and error feedback. One method for assessing an individual's propensity for trial and error learning is through 'foraging' procedures offering uncertain choices between two rewards that dynamically change in their probability of reward. Using foraging procedures, the ability of an individual to learn about their environment has been successfully parameterized with reinforcement learning models (RLM) utilizing the error between predicted and obtained outcomes (Beierholm & Dayan, 2010; Iigaya et al., 2016; Rutledge et al., 2009). It is still uncertain, however, how cues within the individual's environment and drug rewards may modulate the real-time updating of relative value between uncertain options.
Herein, two groups of rats engaged in a foraging task of dynamically changing reward probabilities. Rats initially chose between two nose pokes that probabilistically delivered a sucrose pellet. For the Unsignaled group, choice of either nosepoke resulted in different lever stimuli that were uninformative as to the outcome. For the Signaled group, one nosepoke also resulted in uninformative stimuli, but the informative option produced a lever stimulus only when reward was to follow, while a white cue light indicated reward omission. Following sufficient training, varying doses of remifentanil, a short acting Mu agonist, replaced one choice in the Unsignaled group and the informative choice for the Signaled group. Results revealed relative preferences were sensitive to both Signaled conditions and remifentanil dose. The informative cue in the Signaled group further modulated risk preferences and showed dissociable patterns of behavior relative to the Unsignaled group. Finally, RLM indices successfully parameterized both remifentanil dose-dependent and stimulus signaling effects on trial-by-trial value updating. These results suggest that learning and risk preferences are modulated by environmental cues and drug rewards, and these modulations can be successfully parameterized

using an RLM framework to describe real-time changes in relative value under uncertain conditions.

Disclosures: **A.P. Smith:** None. **J. Beckmann:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.07/UU70

Topic: H.01. Animal Cognition and Behavior

Support: American Cancer Society Institutional Research Grant

USF Proposal Enhancement Grant

Title: Increasing dietary choline attenuates persistent spatial memory deficits in female mice exposed to cyclophosphamide and doxorubicin

Authors: B. E. JOHNS, M. FICKEN, M. E. ENGBERG, *L. WECKER, R. M. PHILPOT
Psychiatry & Neurosci., USF Hlth. Morsani Col. of Med., Tampa, FL

Abstract: Chemotherapy-related cognitive deficits (CRCDs) are a common occurrence for cancer patients. These deficits often persist following treatment and can adversely impact quality of life for decades. To date, the mechanisms mediating the occurrence of CRCDs are unclear and there are no approved treatments for this condition. Given the role of acetylcholine (ACh) in cognitive processes, it is plausible that CRCDs involve a reduction in cholinergic function. Indeed, increasing cholinergic activity with acetylcholinesterase inhibitors attenuates chemotherapy-induced cognitive impairments in mice, but these compounds lead to many adverse effects. Choline (Ch) is a dietary nutrient necessary for the synthesis of ACh, and choline supplementation (+Ch) maintains cholinergic function under conditions of high neuronal demand. Studies have shown that +Ch improves cognitive function in impaired animals and in humans with poor memory, vascular dementia or Alzheimer's disease. Therefore +Ch might be useful for CRCDs. To determine whether +Ch can attenuate spatial memory deficits induced by the chemotherapeutic agents cyclophosphamide (CYP) and doxorubicin (DOX), female BALB/C mice, 64 days of age, were trained in the Morris water maze and baseline performance determined on day 6. Following baseline assessment, mice were placed on a high choline diet (2.0% Ch; Teklad) or remained on standard diet (SD; 0.12% Ch; Teklad). Mice received intravenous injections of CYP (25mg/kg) and DOX (2.5mg/kg) on days 7, 14, 21 and 28 and spatial memory was assessed weekly, from day 13 to 62. On day 6, the % of entries (%E), % of movement (%M) and time (T), in the correct zone exhibited by mice during the probe trial were significantly greater than chance, indicating memory of the platform location. Both groups

exhibited reductions in %E, %M and T on days 13-27 during CYP+DOX exposure. On days 35-62, the performance of +Ch mice was significantly different from SD mice, with +Ch mice exhibiting 8-10% greater %E, 6-20% greater %M and 9-15sec longer T. These data indicate that CYP+DOX administration produces a prolonged impairment in spatial memory and that +Ch can attenuate this deficit when administered during and following chemotherapy. Although it remains to be determined whether this effect extends to other cognitive domains and whether +Ch is prophylactic or therapeutic, these findings suggest that +Ch may be an effective intervention for CRCDs.

Disclosures: B.E. Johns: None. **M. Ficken:** None. **M.E. Engberg:** None. **L. Wecker:** None. **R.M. Philpot:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.08/UU71

Topic: H.01. Animal Cognition and Behavior

Support: FAPESP CEUA-P 2013/12

Title: Towards revealing avoidance test-the role of dopamine

Authors: *G. F. ANTUNES[1], F. V. GOUVEIA[1], M. D. D. SENO[1,2], M. C. DE CARVALHO[2], C. C. DE OLIVEIRA[1], L. C. T. DOS SANTOS[1,4], F. STRAMBIO[1], M. C. DE CASTRO[1,3], M. J. TEIXEIRA[4], J. P. OTOCH[3], M. L. BRANDÃO[2], E. T. FONOFF[4], R. C. R. MARTINEZ[1]
[1]Inst. Sirio Libanês De Ensino E Pesquisa, São Paulo, Brazil; [2]Neurosci. and Behavior, Univ. de São Paulo, Ribeirão Preto, Brazil; [3]Dept. de Técnicas Cirúrgicas, Univ. de São Paulo, São Paulo, Brazil; [4]Dept. de Neurologia, Divisão de Neurocirurgia Funcional, Inst. de Psiquiatria da Univ. de São Paulo, São Paulo, Brazil

Abstract: Avoidance response has been observed in humans and animals. The amygdala, the prefrontal cortex and the dopamine, through the D1 and D2 receptors, are fundamental for modulating this response. There are two population in the avoidance test distinguished by their performance, the good and poor performers (criteria: less than twenty avoidance responses during two consecutive days of training). The objective of our work was to evaluate the role of dopamine as a modulator of this behavior distinction. For that male Wistar rats were used. First, the role of dopaminergic receptors (D1 and D2) was studied in good performers through intraperitoneal injection of saline, D1 antagonist SCH 23390 (0,025 or 0,05 mg/Kg) or D2 sulpiride (20 or 40 mg/Kg). The administration of SCH 23390 reduced the number of avoidance responses ($F_{(4,47)}$=15,10, p&it0.001), concomitantly with a decreased in c-Fos activation only in the amygdala in basolateral (BLA)($F_{(4,18)}$=5,71; p&it0.05) and Medial (Me) nuclei ($F_{(4,20)}$=3,67;

p&it0.05). In the next experiment, good performers received intra-amygdala (basolateral or central nucleus - Ce) administration of D1 SCH23390 antagonist (0,3μg / 0,3μL), D2 sulpiride (0,2 μg / 0,2 μL) or saline; and the poor performers, D1 SKF 38393 agonist (0,4μg / 0,4μL),D2 quinpirole (0,1μg / 0,1μL) or saline. The results showed an increase in avoidance responses after the administration of D2 agonist in BLA ($F_{(2,44)}$=40,46: p<0.001) and Ce nuclei ($F_{(2,94)}$=45,04; p<0.05). The next experiment, evaluated the intrinsic levels of dopamine and its metabolites (DOPAC and HVA). The results showed that good performers had a decreased in DOPAC levels ($F_{(2\ 25)}$=4,35; p&it0.05), and poor performers in HVA lelves ($F_{(2,25)}$=8,18; p&it0.05), both in the right amygdala. The last experiment evaluated the hydroxylase staining comparing good and poor performers considering substantia nigra, ventral tegmental area and locus coeruleus. The results showed an increase in tyrosine hydroxylase positive neurons in good performers considering the substantia nigra ($t_{(1,4)}$ = 4,34; P &it0.05), ventral tegmental area ($t_{1.4)}$ = 10,2, P &it0.001) and locus coeruleus ($t_{(1,3)}$ = 3,55, P &it0.05) as well as a strong correlation between the avoidance behavior equal in substantia nigra (R^2=0.77), ventral tegmental area (R^2= 0.83) and locus coeruleus (R^2=0.89). The main contribution of our work is a better understanding of dopaminergic modulation of avoidance behavior.

Disclosures: G.F. Antunes: None. **F.V. Gouveia:** None. **M.D.D. Seno:** None. **M.C. de Carvalho:** None. **C.C. de Oliveira:** None. **L.C.T. dos Santos:** None. **F. Strambio:** None. **M.C. de Castro:** None. **M.J. Teixeira:** None. **J.P. Otoch:** None. **M.L. Brandão:** None. **E.T. Fonoff:** None. **R.C.R. Martinez:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.09/UU72

Topic: H.01. Animal Cognition and Behavior

Support: NIH Grant 5R01MH097695

Title: Intermittent stimulation of the Nucleus Basalis of Meynert improves performance on the Continuous Performance Task in adult monkeys

Authors: R. LIU[1], J. CRAWFORD[1], P. CALLAHAN[2], A. V. TERRY, JR[3], C. CONSTANTINIDIS[4], *D. T. BLAKE[5]
[1]Brain/Behavior Discovery Inst., [2]Pharmacology/Toxicology, [3]Dept Pharmacol Toxicol, Augusta Univ., Augusta, GA; [4]Wake Forest Univ. Sch. of Med., Winston Salem, NC; [5]Brain and Behavior Discovery Inst., Med. Coll Georgia/Augusta Univ., Augusta, GA

Abstract: Cholinergic transmission has been implicated in working memory and executive function and its loss leads to cognitive impairments. We tested whether cholinergic

pharmacological administration, or electrical activation of the Nucleus Basalis of Meynert, would improve executive function in young adult monkeys during a Continuous Performance Task. In this task, each 500 msec, a square was presented at the center of the screen, with a pseudorandom color chosen from Blue, Red, Yellow, Green and White. The animals were rewarded only if they touched the white square within 500 msec. Stimulation electrodes were implanted in the Nucleus Basalis bilaterally. During stimulation they delivered 60 pulses per second for 20 seconds of each minute. In the pharmacology experiments, we used a cholinesterase inhibitor, donepezil (200ug/kg), a muscarinic cholinergic antagonist, scopolamine (6.25ug/kg) and a nicotinic cholinergic inhibitor, mecamylamine (300ug/kg) to investigate the effects on animals' performance. Deep brain stimulation in Nucleus Basalis improved performance in two animals. Percent correct touches increased from 67.1% to 70.5% (Animal One) and 81.6% to 84.2% (Animal Two, binomial test $p<0.001$ for both monkeys), respectively. Unexpectedly, donepezil application alone, decreased animals performance from 67.1% to 64.4% and 81.6% to 77.8%, respectively ($p<0.001$ for both monkeys). When electrical stimulation and donepezil were combined, percent correct decreased from 74.6% to 71.6% and 88.2% to 84.5%, respectively ($p<0.01$ for both monkeys). To investigate cholinergic receptor subtype impact on performance, we used scopolamine or mecamylamine. Scopolamine decreased the animals' percentage of correct responses (animal 1: 54.8% to 50.1%, $p<0.01$ animal 2: 76.9% to 39.9%, $p<0.001$) while mecamylamine increased it (animal 1: 58% to 65.7%,$p<0.01$, animal 2: 76.9% to 81.2%,$p<0.001$). The results indicate that deep brain stimulation in Nucleus Basalis is superior to donepezil, a frontline medication used to improve cholinergic action, in increasing cognitive performance in a sustained attention task. The improvement is likely the result of complex interactions between projecting cell types from Nucleus Basalis and cholinergic receptor subtypes, as well as interplay between differing roles of acetylcholine in the striatum and in thalamocortical networks.

Disclosures: R. Liu: None. **J. Crawford:** None. **P. Callahan:** None. **A.V. Terry:** None. **C. Constantinidis:** None. **D.T. Blake:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.10/UU73

Topic: H.01. Animal Cognition and Behavior

Support: FAPESP

CNPq

CAPES

FAEPA

Title: Medial prefrontal cortex glutamate and canabinoide systems modulating reconsolidation and extinction of contextual fear conditioning memory

Authors: *L. B. RESSTEL[1], D. G. REIS[2], S. F. LISBOA[3]
[1]Schl Med, FMRP-USP, Ribeirao Preto- SP, Brazil; [2]Univ. Sao Paulo, Ribeirao Preto, Brazil; [3]Pharmacol., Univ. of São Paulo - FMRP, Ribeirao Preto, Brazil

Abstract: The ventral portion of medial prefrontal cortex (vMPFC) is involved in the modulation of processes involving the expression and extinction of conditioned emotional response (CER). vMPFC NMDA and CB1 receptors present opposing roles on behavioral (Freezing) and autonomic modulation (mean arterial pressure (MAP), heart rate (HR) and cutaneous temperature (TC), during expression of the CER. However, the role of a possible interaction between these receptors on autonomic and behavior responses associated to reconsolidation and extinction of contextual fear memories has not be fully studied. The aim of the present study was to investigate the effect of antagonism of vMPFC NMDA and CB1 receptors on reconsolidation and extinction processes. Male Wistar rats (250g) had a radio-telemetry probe implanted for recording autonomic activity and guide cannulas in vMPFC for drug administration. Day 1: Preconditioning started 1 wk after guide cannula implantation and consisted of one 10-min-long pre-exposure (habituation) in the footshock box. After four hours, the animals received 4 electrical footshock (0.8 mA, 1 s). Day 2: Two groups were tested, reconsolidation group (5 min session) or extinction group (30 min session) re-exposure without footshock presentation. Each group received two intra vMPFC injections (1st vehicle or CB1 antagonist AM251 30 pmol / 2nd vehicle or NMDA antagonist LY235959 0.4 nmol) 15 or 10 min before reconsolidation or extinction sessions forming 4 groups (Vehicle x Vehicle / Vehicle x LY235959 / AM251 x Vehicle / AM251 x LY235959). Day 3: The test session take place in the next day consisting of a 10-min re-exposure to the footshock box without any shock or injection given. Reconsolidation Day 2: No significant differences were observed between all groups on CER (Freezing: $F_{(3, 19)}$=0.4; P>0.05 ΔMAP: $F_{(3, 85)}$ = 1.386 P<0.05 ΔHR: $F_{(3, 85)}$ = 0.7 P<0.05 ΔCT: $F_{(3, 85)}$=22; P<0.05). Day 3: Group AM251 30 pmol x vehicle presented an elevated expression of CER compared to all other groups (Freezing: $F_{(3,19)}$=8.9; P<0.05 ΔMAP: $F_{3,85}$=13 P<0.05 ΔHR: $F_{3,85}$=42; P<0.05 ΔCT: $F_{3,85}$=22; P<0.05). Extinction Day 2: No significant difference were observed between all groups on CER (Freezing: $F_{(3, 119)}$ = 0.5; P>0.05 ΔMAP: $F_{(3, 102)}$ = 1,1 P>0.05 ΔHR: $F_{(3, 102)}$ = 0.6 P>0.05 ΔCT: $F_{(3, 102)}$ = 0.3 P>0.05. Day 3: Group AM251 30 pmol x vehicle presented an elevated expression of compared to all other groups (Freezing: $F_{(3,19)}$=8.9; P<0.05 ΔMAP: $F_{3,85}$=6.37 P<0.05 ΔHR: $F_{3,85}$=19; P<0.05 ΔCT: $F_{3,85}$=31; P<0.05). The interaction between NMDA and CB1 receptors in vMPFC modulates both reconsolidation and extinction process associated to contextual fear memories

Disclosures: L.B. Resstel: None. **D.G. Reis:** None. **S.F. Lisboa:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.11/UU74

Topic: H.01. Animal Cognition and Behavior

Support: JSPS Grant 16K04419

Title: Spontaneous novel object recognition, object location recognition, and temporal order discrimination in rats neonatally treated chronically with MK-801

Authors: *T. HATAKEYAMA[1], H. FURUIE[2,3], K. YAMADA[1], Y. ICHITANI[1]
[1]Lab. of Behavioral Neurosci., Univ. of Tsukuba, Tsukuba-Shi, Japan; [2]Ctr. for Med. Sci., Ibaraki Prefectural Univ. of Hlth. Sci., Ami, Japan; [3]Natl. Inst. of Mental Hlth., Natl. Ctr. of Neurol. and Psychiatry, Tokyo, Japan

Abstract: Glutamatergic N-methyl-D-aspartate (NMDA) receptors play a critical role in early brain development. Rats chronically treated with a NMDA antagonist in early developmental stage have been reported to show spontaneous object recognition impairment in adulthood, suggesting the usefulness of neonatal NMDA receptor blockade as a potential model for schizophrenia. Previous studies, however, focused mainly on novel object recognition, and little is known whether the deficits in the spontaneous object location and temporal order recognitions would appear in adulthood. Therefore, here we investigated the effects on the two types of tests as well as novel object recognition in rats neonatally treated chronically with MK-801.
Male Wistar rat pups received injection of 0.4 mg/kg of MK-801 or vehicle (SAL) twice a day on postnatal day 7-20. At the age of 10-13 weeks, three types of spontaneous object recognition tests were conducted. Firstly, the novel object recognition test consisted of a sample phase (5 min) in which rats were exposed to a pair of identical objects (AA) in an arena, a delay of 10 min, and a test phase (5 min) in which one of the two objects was replaced by a novel object (AB) and preference was assessed by comparing the time spent exploring the familiar versus novel object. Secondly, the object location recognition test consisted of a sample phase (10 min) in which two identical objects (CC) were placed, a delay of 10 min, and a test phase (5 min) in which one object was moved to a new position in the arena and novel location preference was assessed. Thirdly, the temporal order recognition test consisted of a sample phase (5 min) in which the first presentation was a pair of identical objects (DD), and then, 125 min later, the second presentation was another pair of objects (EE), a delay of 10 min, and a test phase in which two different objects (DE) were placed and old object preference was assessed by comparing the time spent exploring the old versus recent object.
In the novel object recognition test, there was not a significant difference of discrimination ratio (DR) between MK-801- and SAL-treated groups. However, if the delay was prolonged to 60

min, MK-801 group tended to show a lower DR compared with SAL group. In the object location recognition test, MK-801 group tended to show a lower DR compared with SAL group. In the temporal order recognition test, there was a significant difference between neonatal treatment groups, and the DR of MK-801 rats was significantly lower than that of SAL rats. These results suggest that chronic neonatal NMDA receptor blockade may induce long-lasting effects on the brain regions subserving temporal order discrimination in adulthood.

Disclosures: T. Hatakeyama: None. **H. Furuie:** None. **K. Yamada:** None. **Y. Ichitani:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.12/UU75

Topic: H.01. Animal Cognition and Behavior

Support: R01MH098985

R21AG044530

R21AG051753

CA200417

DA024628

Title: The chemotherapeutic agent paclitaxel selectively impairs reversal learning while sparing prior learning, new learning and episodic memory

Authors: *D. PANOZ-BROWN, L. M. CAREY, A. E. SMITH, M. GENTRY, C. M. SLUKA, H. E. CORBIN, J.-E. WU, A. G. HOHMANN, J. D. CRYSTAL
Psychology, Indiana Univ. Bloomington, Bloomington, IN

Abstract: Chemotherapy is widely used to treat patients with systemic cancer. The efficacy of cancer therapies is frequently undermined by adverse side effects that have a negative impact on the quality of life of cancer survivors. Cancer patients who receive chemotherapy often experience chemotherapy-induced cognitive impairment across a variety of domains including learning, memory, and attention. In the current study, the impact of paclitaxel, a taxane derived chemotherapeutic agent, on episodic memory, prior learning, new learning, and reversal learning were evaluated in rats. Neurogenesis was quantified post-treatment in the dentate gyrus of the same rats using immunostaining for Bromodeoxyuridine (BrdU) and Ki67. Paclitaxel treatment selectively impaired reversal learning while sparing prior learning, new learning, and episodic memory. Furthermore, paclitaxel-treated rats showed decreases in markers of hippocampal cell

proliferation, as measured by markers of cell proliferation assessed using Ki67 and BrdU immunostaining. This work highlights the importance of using multiple measures of learning and memory to identify the pattern of impaired and spared aspects of chemotherapy-induced cognitive impairment.

Disclosures: D. Panoz-Brown: None. **L.M. Carey:** None. **A.E. Smith:** None. **M. Gentry:** None. **C.M. Sluka:** None. **H.E. Corbin:** None. **J. Wu:** None. **A.G. Hohmann:** None. **J.D. Crystal:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.13/UU76

Topic: H.01. Animal Cognition and Behavior

Support: Pfizer

Memory Pharmaceuticals

Prime Behavior Testing Laboratories

NIDA DA029127

Title: Atomoxetine enhances working memory and other behavioral domains in young and aged animals

Authors: *A. V. TERRY, JR[1], P. M. CALLAHAN[1], M. PLAGENHOEF[1], D. BLAKE[2]
[1]Dept Pharmacol Toxicol, [2]Brain Behavior Discovery Inst. & Dept. of Neurol., Augusta Univ., Augusta, GA

Abstract: Atomoxetine is a norepinephrine reuptake inhibitor that is approved for the treatment of attention deficit hyperactivity disorder (ADHD) in children, adolescents, and adults. While it is typically considered a second line therapy for ADHD when compared to stimulants such as methylphenidate and amphetamine, it offers the advantages of reduced abuse liability, decreased risk of motor side effects and it can serve as an alternative medication for patients who are not responsive to stimulants. While there is some evidence that atomoxetine (in addition to its positive effects on attention) may have the potential to improve other domains of cognition such as working memory, this subject has not been systematically studied and, moreover, atomoxetine has not been studied extensively in aged subjects. In the experiments described here, we evaluated atomoxetine in doses ranging from 0.3-5.6 mg/kg (i.p. 30 or 60 minutes before behavioral testing) in adult rats in a variable inter-trial interval (VITI) version of a five choice serial reaction time task (5C-SRTT) and a radial arm maze (RAM) delayed non-match to

position (DNMTP) task. Atomoxetine, doses ranging from 0.03-1.0 mg/kg (i.m., 60 minutes before behavioral testing) was also evaluated in a distractor version of the delayed match to sample task (DMTS-D) in aged (greater than 20 years old) monkeys. In rats, atomoxetine was associated with dose-dependent improvements of inhibitory response control in the 5C-SRTT, as well as with improved performance of the RAM-DNMTP task. In aged monkeys, atomoxetine was associated with dose-dependent decreases in distractibility in the DMTS-D task. Collectively, these studies support the argument that atomoxetine has the potential to decrease distractibility and to improve certain domains of cognition (e.g., working/short memory) and as well as to improve impulse control. Each of these positive findings has relevance to neuropsychiatric disorders such as ADHD and schizophrenia as well as age-related disorders such as Mild Cognitive Impairment (MCI) and Alzheimer's disease.

Disclosures: A.V. Terry: None. **P.M. Callahan:** None. **M. Plagenhoef:** None. **D. Blake:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.14/UU77

Topic: H.01. Animal Cognition and Behavior

Support: High Point University Research Advancement

Title: Selective D4R antagonist ligands as molecular tools to study addiction

Authors: *C. A. BOATENG[1], M. M. DAY[2], R. B. FREE[2], D. R. SIBLEY[2], K. STEWART[1], T. M. KECK[3]
[1]Basic Pharmaceut. Sci., High Point University, Fred Wilson Sch. of Pharm, High Point, NC; [2]Mol. Neuropharm. Section, Natl. Inst. of Neurolog. Disorders and Stroke, Bethesda, MD; [3]Chem. & Biochem., Rowan Univ., Glassboro, NJ

Abstract: Dopamine D4 receptors (D_4R) are G protein-coupled receptors predominantly expressed in the prefrontal cortex where they play an important role in cognition, attention, and decision making. Previous studies using D_4R ligands of varying efficacies have determined that D_4R signaling alters behavior in animal models of drug addiction and cognition. Developing novel D_4R-selective ligands will allow more detailed investigations into the biological role of D_4R signaling in the brain and assist in medication development for neuropsychiatric disorders, including Alzheimer's disease and substance use disorders (SUD). In order to develop new candidate medications for SUD, we have designed, synthesized, and pharmacologically evaluated novel D_4R antagonist ligands. Starting with the D_4R antagonist 2-(4-(4-(pyrimidin-2-yl)piperazin-1-yl)propyl)benzo [*d*]thiazole as our parent compound, we optimized a next-

generation compound library by using a computational modelling approach. We hypothesized that structural modifications of the parent compound template would produce novel ligands with high D_4R binding affinity, receptor subtype selectivity, and efficacy. In this pursuit, we have produced a library of eighteen compounds with varied substitutions on the pyrimidinylpiperidinyl (PP) ring and/or the benzothiazole moieties. These novel ligands were synthesized and their *in vitro* binding affinities were determined using [^{3}H]*N*-methylspiperone radioligand binding in HEK293 cells expressing dopamine D2-like receptors. By modifying the pyrimidinylpiperidinyl and benzothiazole moieties, we have identified several high-affinity compounds ($K_i \leq 4.85$ nM) with >100-fold selectivity at the D_4R versus D_2 and D_3 receptors. Based on binding profiles, a subset of analogues were evaluated in functional assays measuring β-arrestin recruitment and cAMP production. These new lead compounds will be further evaluated for effects in animal models of cognition and/or drug addiction.

Disclosures: C.A. Boateng: None. **M.M. Day:** None. **R.B. Free:** None. **D.R. Sibley:** None. **K. Stewart:** None. **T.M. Keck:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.15/UU78

Topic: H.01. Animal Cognition and Behavior

Support: American Cancer Society USF Institutional Research Grant

USF Proposal Enhancement Grant

Title: Acetycholinesterase inhibitors prevent, but do not reverse, persistent spatial memory deficits induced in female mice by cyclophosphamide and doxorubicin exposure

Authors: M. FICKEN, B. E. JOHNS, M. E. ENGBERG, L. WECKER, *R. M. PHILPOT
Psychiatry and Behavioral Med., Univ. South Florida, Morsani Col. of Med., Tampa, FL

Abstract: Cancer patients often experience impairments in cognitive function during and following chemotherapy. These chemotherapy-related cognitive deficits (CRCDs) can persist for decades, impairing day-to-day functioning and adversely impacting quality of life. Acetylcholinesterase (AChE) inhibitors improve cognitive function in individuals suffering from Alzheimer's disease and research in mice indicates that the AChE inhibitor donepezil (DON) may be beneficial for the prevention or treatment of CRCDs induced by methotrexate and 5-fluorouracil. To determine if DON can attenuate CRCDs induced by cyclophosphamide (CYP) and doxorubicin (DOX), if these actions are prophylactic or therapeutic, and if the AChE inhibitor galantamine (GAL) produces similar effects, female BALB/C mice, 64 days of age,

were trained in the Morris water maze. Baseline performance was determined on day 6 and mice were assessed weekly from day 13 to 62. Mice received weekly intravenous injections of CYP (25mg/kg) and DOX (2.5mg/kg) on days 7, 14, 21 and 28. For exp 1 and 2, CYP+DOX administration reduced the % of entries (%E), % of movement (%M), and time spent (T) in the correct zone during the probe trial, indicating impaired spatial memory. On day 35, 7 days following the final injection of CYP+DOX, mice began receiving daily injections (s.c.) of DON (3mg/kg), GAL (3mg/kg), or equivalent volumes saline (SAL). DON- and GAL-injected mice did not differ from SAL mice and did not improve on any measure of spatial memory during treatment (days 35-62). For exp 3, mice received daily injections (s.c.) of DON (3mg/kg), GAL (3mg/kg) or SAL during the 4 weeks of CYP+DOX administration, from day 7-34. Relative to day 6, the %E, %M, and T exhibited by mice was reduced by 13-38%, 14-58% and 19-56%, respectively, on days 13-34. On days 41-62, the performance of DON- and GAL-injected mice was significantly greater than SAL-injected mice, with DON- and GAL mice exhibiting 11-25% greater %E, 7-28% greater %M and 11-14sec longer T in the correct zone than SAL mice. The performance of DON and GAL mice did not differ from each other on any measure. The results indicate that AChE inhibitors do not reverse spatial memory deficits induced by exposure to CYP+DOX, but can attenuate deficits if administered during CYP+DOX exposure. These findings suggest that AChE inhibitors may be useful for the prevention of CRCDs if administered concurrent with chemotherapy.

Disclosures: M. Ficken: None. **B.E. Johns:** None. **M.E. Engberg:** None. **L. Wecker:** None. **R.M. Philpot:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.16/UU79

Topic: H.01. Animal Cognition and Behavior

Title: Effects of heat shock protein induction on spatial learning and memory in mice

Authors: *R. TANOUE, Y. MORITA, S. YAMAMOTO, N. HASHIKAWA-HOBARA, N. HASHIKAWA
Okayama Univ. of Sci., Okayama-Shi, Japan

Abstract: Learning and memory functions in organisms are important functions directly connected to life support such as food intake and danger avoidance, and there are many similar mechanisms from lower organisms to higher organisms. Heat shock protein (HSP) is a group of proteins expressed when cells undergo stress such as heat, ultraviolet rays, burns etc, and acts as a molecular chaperone that folds nascent proteins and repairs denatured proteins. HSP expression is closely related to diseases of the central nervous system, but its effect on learning and memory

is not clear. In the present study, to clarify the role of HSP in learning and memory, geranyl-geranyl acetone (GGA) known as HSP inducer was administered orally to C57BL6J mice, and assessed Morris water maze test (spatial memory), Rota-rod test (motor learning memory), Passive avoidance test (fear memory), and Y maze test (working memory). Furthermore, the level of HSP mRNA in the brain was analyzed by real-time PCR. In the Rota-rod test, the Passive avoidance test, and the Y maze test, GGA administered did not show any significant differences when compared to control. However, GGA treatment mice learned the water maze task at a higher rate than control and had better performance scores by the end of the training period and on a probe trial. Our findings demonstrated that GGA treatment lead to ameliorate spatial memory.

Disclosures: **R. Tanoue:** None. **Y. Morita:** None. **S. Yamamoto:** None. **N. Hashikawa-Hobara:** None. **N. Hashikawa:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.17/UU80

Topic: H.01. Animal Cognition and Behavior

Support: NIH Grant DA029252

Title: An automated test of olfactory working memory in rats: Effects of MK-801

Authors: H. A. KELLIHER[1], S. A. NELSON[2], K. DYER[2], S. ACCATTATO[2], L. RICHARDSON[2], M. MATHEWS[2], *J. GALIZIO[2]
[1]Psychology, [2]Univ. of North Carolina at Wilmington, Wilmington, NC

Abstract: Available procedures to study working memory capacity in rodents generally use manual arena or maze tasks. For example, the odor span task uses an incrementing non-match to sample procedure in which digging in cups scented with novel olfactory stimuli results in reinforcement on each trial, while responses to previously presented stimuli are not reinforced. The present study modified this procedure for use in an automated operant chamber using a 15-channel olfactometer. Rats were trained on a go, no-go procedure to make nose-poke responses in a port through which odorants were delivered. A series of odorants was presented several times during a session. Responses to each odorant were reinforced on an FI 5-s schedule the first time it was presented, but once an odor had been presented, responses to that odor were no longer reinforced. Rats rapidly learned to differentiate between novel and familiar odors and responded at high rates to new stimuli and much lower rates to repeated stimuli. Accuracy was disrupted with the insertion of a mid-session distractor task. The effects of NMDA antagonist, MK-801 were assessed in six rats and produced a dose-dependent impairment in accuracy. These

results were similar to findings obtained in the odor span task, and illustrate the value of studying memory for multiple stimuli using an automated procedure.

Disclosures: H.A. Kelliher: None. **S.A. Nelson:** None. **K. Dyer:** None. **S. Accattato:** None. **L. Richardson:** None. **M. Mathews:** None. **J. Galizio:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.18/UU81

Topic: H.01. Animal Cognition and Behavior

Support: CIHR fund (MOP-102482)

Title: Nicotinergic alpha7 and alpha4/beta2 sub-receptor agonists differentially enhance reversal learning speed and attentional filtering of distraction in a non-human primate model

Authors: *M. AZIMI[1], M. OEMISCH[1], T. WOMELSDORF[1,2]
[1]Biol., York Univ., Toronto, ON, Canada; [2]Psychology, Vanderbilt Univ., Nashville, TN

Abstract: Nicotinic acetylcholine receptors (nAChR) are believed to modulate attention, memory, and higher executive functioning through the alpha 4-beta 2 and/or the alpha 7 nACh sub-receptors. These suggestions are mainly derived from rodent studies using task paradigms with only an indirect relationship to flexible attention and learning behavior of the primate behavioral repertoire. We therefore set out to identify the specific contributions of two highly selective agonists for alpha7 (α7) and alpha4-beta2 (α4β2) nAChRs in mediating flexible reversal learning and attentional filtering of distractors in a non-human primate model. We used two different doses of each agonist to compare the effects of drug to non-drug conditions while the monkey performed a feature-based reversal learning task. The task allowed dissociating subcomponent processes of attentional filtering, learning efficacy, motivation, impulsivity and perseveration tendencies.
We found for both sub-receptor agonists a dose-dependent enhancement of specific aspects of reversal performance. The optimal dose for α7 increased performance accuracy and sped up learning in reversals of the rewarded feature value. However, we found that this effect was specific to the first 25 minutes after the task started. This duration of behavioral improvement corresponded to the period when the agent was at its peak concentration in blood plasma. In contrast to the α7 effect, the best dose of the α4β2 nAChR agonist increased performance accuracy specifically in those subsets of trials that had an enhanced demand to filter out distracting visual information. However, in contrast to these behaviorally enhancing effects on reversal learning speed (α7) and attentional filtering (α4β2), neither drug modulated motivation, impulsivity, or perseveration rates.

In summary, our results document sub-receptor specific effects on higher order learning and attention functions in a healthy non-human primate. The effectiveness of both agonists was dependent on the inferred, relative concentration of the drug in blood plasma. These findings are an important first step to identify the neuromodulatory mechanisms of attention and learning functions in the primate brain. We speculate that these results could be a first step towards the development of function-specific drugs alleviating cognitive deficits evident in multiple neuro-psychiatric diseases.

Disclosures: M. Azimi: None. **M. Oemisch:** None. **T. Womelsdorf:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.19/UU82

Topic: H.01. Animal Cognition and Behavior

Support: ERC-AG / FP7-IDEAS-ERC

Title: Behavioral flexibility is improved by chronic fluoxetine treatment through BDNF/TrkB signaling

Authors: *J. UMEMORI[1], G. DIDIO[1], F. WINKEL[1], H. ANTILA[2], M. L. POU[3], V. VÕIKAR[1], R. GURADO[1], E. CASTREN[2]
[1]Neurosci. center, [2]Neurosci. Ctr., Univ. of Helsinki, Helsinki, Finland; [3]Biomed. Science,, Univ. of Barcelona, Barcelona, Spain

Abstract: Brain-derived neurotrophic factor (BDNF) is a major regulator of neuronal plasticity through the activation of its receptor tyrosine kinase TrkB. We have demonstrated that chronic fluoxetine treatment combined with fear extinction or monocular deprivation, but neither treatment alone, induced an enduring loss of conditioned fear memory or shift of ocular dominance in the visual cortex, respectively, activated by BDNF/TrkB pathway (Vetencourt et al., 2008; Karpova et al., 2011). Thus, chronic antidepressant drug treatment induces a critical period-like plasticity, which allows brain networks to better adapt to the internal and external milieu in the adult brain. However it is not clear whether spatial reversal learning or behavioral flexibility is improved by chronic fluoxetine treatment during learning process via BDNF/TrkB pathway.
Here we used BDNF heterozygous knockout (Bdnf hKO) and heterozygous TrkB knockout specifically in Parvalbumin (PV)-expressing interneurons (hPV-TrkB) mice, and assessed the effects of fluoxetine on plasticity using Intellicage (NewBehavior AG, Zurich, Switzerland), where transponder-implanted female mice were group-housed in a genotype-mixed manner. In IntelliCage water-deprived mice were rewarded by access to water when visiting conditioning

corners in defined sequence during the acquisition phase, and then it was exchanged to the opposite direction in the reversal phase. In wild-type mice, chronic treatment with fluoxetine did not affect acquisition of the spatial memory but improved “reversal” learning. Bdnf hKO mice showed impaired acquisition of spatial memory as previously indicated (Linnarsson et al., 1997), but chronic fluoxetine treatment improved spatial memory in the acquisition and reversal phase to similar level as in the wild-type mice. On the other hand, hPV-TrkB mice did not show deficits in learning or memory in the acquisition or reversal phase, but the treatment with fluoxetine did not improve reversal learning as in the wild-type mice. These results indicate that fluoxetine promotes behavioral flexibility in the reversal learning task by activating TrkB signaling in PV interneurons.

Disclosures: **J. Umemori:** None. **G. Didio:** None. **F. Winkel:** None. **H. Antila:** None. **M.L. Pou:** None. **V. Võikar:** None. **R. Gurado:** None. **E. Castren:** None.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.20/UU83

Topic: H.01. Animal Cognition and Behavior

Support: Supported by H. Lundbeck A/S

Title: The phosphodiesterase 1 inhibitor Lu AF64386 increases cGMP and cAMP in the brain and exerts procognitive effects in the rat

Authors: *A. MORK, H. S. LINDGREN, V. NIELSEN, C. T. CHRISTOFFERSEN, J. KEHLER, J. NIELSEN
H. Lundbeck A/S, Copenhagen-Valby, Denmark

Abstract: Phosphodiesterase 1 (PDE1) is a dual substrate enzyme hydrolyzing cGMP and cAMP and thus plays a major role in cell signaling. There are 3 PDE1 genes. PDE1A is expressed at high levels in the brain primarily in the output neurons of layers 5/6 of the cortex and in CA1-3 of the hippocampus. PDE1B is also expressed at high levels in the brain with highest expression in the striatum, but also expressed in the cortex, dentate gyrus and other brain regions. PDE1C is expressed at low levels in the brain. Various preclinical studies have suggested an association between activity inhibition of different PDE families and cognition enhancing actions. In the present study we investigated the effect of the PDE1 inhibitor Lu AF64386 (1) on PDE1 in vivo binding and levels of cGMP and cAMP in the brain of awake rats. Furthermore, the effect of Lu AF64386 was studied in preclinical models of cognitive function. PDE1 occupancy was measured by in vivo binding, i.e., displacement of a [^{3}H]-labelled PDE1 inhibitor after pretreating with un-labelled Lu AF64386. Effects on extracellular levels of cGMP and cAMP in

the brain were measured by microdialysis in freely-moving rats and the content of nucleotides in the microdialysis samples was determined by HPLC and tandem mass spectrometry analysis. Effects on executive function and memory were evaluated by using the attentional set shifting task and contextual fear conditioning. Subcutaneous administration of Lu AF64386 at 0.3-30 mg/kg dose-dependently occupied striatal PDE1 (EC50 2.5 mg/kg), reaching full occupancy at the highest dose. Moreover, Lu AF64386 dose-dependently increased cGMP and cAMP in the rat striatum, the prefrontal cortex and the hippocampus. Extracellular levels of cGMP reached > 700% of baseline for rats treated with the highest dose, whilst cAMP levels within those rats reached 150 - 200% of baseline. Similar doses of Lu AF64386 also improved memory function assessed in the contextual fear conditioning task, as well as attenuating the subchronic phencyclidine-induced deficit at the extradimensional shift in the set-shifting task. Since PDE1 is highly expressed in brain areas mediating cognition, Lu AF64386-induced increases in cyclic nucleotides may affect impaired synaptic function and improve cognitive functions in patients with psychiatric disorders. 1. Quinazolin-THF-amines as PDE1 inhibitors, Kehler J, Kyhn Rasmussen L, Kyhn; Langgaard M; PCT Int. Appl. (2015), WO 2015091805

Disclosures: **A. Mork:** A. Employment/Salary (full or part-time):; H. Lundbeck A/S. **H.S. Lindgren:** A. Employment/Salary (full or part-time):; H. Lundbeck A/S. **V. Nielsen:** A. Employment/Salary (full or part-time):; H. Lundbeck A/S. **C.T. Christoffersen:** A. Employment/Salary (full or part-time):; H. Lundbeck A/S. **J. Kehler:** A. Employment/Salary (full or part-time):; H. Lundbeck A/S. **J. Nielsen:** A. Employment/Salary (full or part-time):; H. Lundbeck A/S.

Poster

429. Learning and Memory: Pharmacology

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 429.21/VV1

Topic: H.01. Animal Cognition and Behavior

Support: MEXT of Japan, Grant-in-Aid for Scientific Research (C, #24530917)

Title: The relationship between blood pressure reduction and working memory performance in spontaneously hypertensive rats (SHR): Understanding the effects of hydralazine and strains differences in rats

Authors: *T. SATO
Social Welfare, Nagano Univ., Ueda, Japan

Abstract: Spontaneously hypertensive rats (SHRs) were originally developed as a model to study hypertension in humans. It is well known that SHRs exhibit certain behavioral characteristics, such as, increased locomotor activity, which decreases with decreasing blood

pressure (BP) induced by the intravenous administration of hypotensive drugs. However, it is unclear if the cognitive functions of SHRs, such as learning and memory, change upon reducing BP. This study aimed to determine the effects of BP reduction on SHR memory function. For this purpose, we used 8 well-trained SHRs, along with 8 Wistar-Kyoto strain (WKY) rats as normotensive control, for a delayed-matching-to-place (DMTP) task, results of which were compared between the two rat-strain groups in terms of the number of correct responses. Upon the completion of pre-training, all rats were repeatedly tested in the DMTP task four times. In each session, rats were intravenously injected with a vasodilator, hydralazine, at one of the four different doses (0, 0.1, 0.3, 0.6 mg/kg) in a randomized order. During the DMTP task, rats were reinforced with food when they made a matching response, i.e., when the rat pressed the retractable lever inserted as the sample at the beginning of the respective trial. In addition, each task session had 100 trials. At the beginning of each trial, one lever was randomly selected as the sample and inserted in the chamber. When the rat pressed the lever, the lever retracted and the food-cup light in the rear wall illuminated. After the rat made a nose poke to the food cup twice, both levers were inserted. Both levers retracted when one of them was pressed. To obtain a retention gradient, the time interval between the first nose-poke and the end of the delay interval, wherein no nose-poke was effective for the presentation of both levers, was varied amongst five different time lengths (0, 2, 5, 10, 20 s) in a randomized order. A three-factor ANOVA and its subsequent analyses indicated that, when the delay time was more than 2 s, the number of correct responses gradually decreased in proportion to the delay time length. Subsequent analysis also revealed that the number of correct responses following the injection of 0.3 and 0.6 mg/kg hydralazine was smaller than that after saline injection; however, the size of the difference between these dosages was relatively small. Additionally, the strain difference between SHR and WKY rats was not statistically significant. In conclusion, these results reveal that the effects of average to high doses of hydralazine on memory performance in the DMTP task are statistically significant, though relatively small, and the strain difference between SHR and WKY rats is not evident.

Disclosures: T. Sato: None.

Poster

430. Neuromodulation and Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 430.01/VV2

Topic: H.02. Human Cognition and Behavior

Support: NIH Grant MH63901

NIH Grant NS40813

Title: Modulating the cortico-striatal output gate of working memory

Authors: *J. MILLER[1], A. KIYONAGA[1], R. B. IVRY[1,2], M. D'ESPOSITO[1,2]
[1]Helen Wills Neurosci. Inst., [2]Dept. of Psychology, UC Berkeley, Berkeley, CA

Abstract: Working memory (WM) confers the unique ability to guide behavior based on internal representations, and to therefore plan for the near future. However, some WM representations are irrelevant to current goals. For instance, WM content is often maintained for a future goal, during performance of more immediate tasks. During WM maintenance, WM content can bias perception in favor of similar stimuli. The relevance of the WM content to the immediate task may determine its activation status in either a prioritized or latent state (i.e., 'silent coding'; Stokes, 2015), as well as the extent of its influence on current perception. However, it is unknown whether WM content has an analogous impact on the execution of actions: How and when does active thought content involuntarily influence motor behavior? A 'gating' process has been proposed to describe goal-dependent, flexible control over what information gets entered into WM (i.e., input gating), and when those WM representations are selected to control behavior (i.e., output gating; Chatham & Badre, 2015). Prefrontal-striatal circuitry has been implicated in the control of this gating process, but there is very little empirical evidence for the output gating hypothesis. Here, we sought to (1) behaviorally engage the putative WM output gating mechanism, and (2) causally test the cortico-striatal circuitry of WM output gating through transcranial magnetic stimulation (TMS). We developed a novel behavioral task to test whether WM content can involuntarily influence behavior, similar to how it biases perception: participants maintained a WM sample (either the word "up" "down" "left" or "right") and then performed a motor reaching task to a cued location during the WM delay. WM content could be compatible or incompatible with the goals of the reaching task. Movement initiation was fastest when WM content was compatible and slower when WM content was incompatible with the action goal, suggesting that WM content can influence response initiation for a different task. This compatibility effect was only evident, however, when the movement task goal was indicated with a symbolic cue; there was no compatibility effect when the movement goal was exogenously cued, suggesting the WM content influenced response selection rather than implementation. Further, TMS delivered to lateral prefrontal cortex (PFC)—targeted based on striatal connectivity—modulated the magnitude of the behavioral effect. Our results suggest that active WM content can involuntarily influence actions, and that the selection of representations from within WM to guide behavior (i.e., output gating) is controlled by prefrontal-striatal interactions.

Disclosures: J. Miller: None. **A. Kiyonaga:** None. **R.B. Ivry:** None. **M. D'Esposito:** None.

Poster

430. Neuromodulation and Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 430.02/VV3

Topic: H.02. Human Cognition and Behavior

Support: NIH MH63901

NIH MH103842

NSF DGE 1106400

Title: Reactivation and suppression of representations in working memory using frequency specific tms

Authors: *J. RIDDLE[1], D. CELLIER[3], S. DHANANI[3], M. DESPOSITO[2]
[2]Helen Wills Neurosci. Inst., [1]Univ. of California Berkeley, Berkeley, CA; [3]Univ. of California, Berkeley, Berkeley, CA

Abstract: Working memory (WM) is the ability to maintain and manipulate information after it is no longer present in the environment. A retro-cue during the delay period of a WM task triggers reactivation of a subset of WM representations, providing a useful method for dissociating WM encoding from WM reactivation processes. Magnetoencephalography studies have shown that theta frequency oscillations emerge in areas that maintain WM representations during reactivation with a retro-cue (Wallis et al. 2016), whereas when WM representations are inhibited or suppressed, alpha frequency oscillations emerge (Sauseng et al. 2009).To establish the necessity of neural oscillations in WM, we used rhythmic transcranial magnetic stimulation (TMS) to causally manipulate alpha and theta oscillations during the reactivation and suppression of WM traces. Subjects were presented with 4 colored squares in each visual field. After a 1 second delay, a retro-cue (arrow pointing to one side) instructed subjects to remember the items in one visual field and the items in the other visual field could be forgotten. After another 1 second delay, subjects judged an array of 4 colored squares to be a match or non-match to the memory array. Subjects were screened for their ability to utilize the retro-cue over a non-informative neutral cue. In another session, 14 screened subjects performed the task during fMRI for functional localization of the left middle frontal gyrus (L-MFG) and left inferior intraparietal sulcus (L-IPS). In the 3rd and 4th sessions, subjects received online TMS to either L-MFG or L-IPS immediately after the presentation of the retro-cue. The key manipulation was the TMS frequency: theta (5 Hz), alpha (10 Hz), or arrhythmic control. Our hypothesis was that disruption of theta frequency oscillations would impact the retro-cue benefit of items in the right visual field, which was confirmed as only right retro-cues during theta TMS to L-IPS and L-MFG decreased the retro-cue benefit relative to arrhythmic control. We also hypothesized that entrainment of alpha frequency oscillations would boost suppression of items in the right visual field, improving the left-visual field retro-cue benefit. This effect was not significant but the retro-cue accuracy benefit in the left visual field following alpha TMS was inversely related to the accuracy detriment in the right visual field that was found following theta TMS.These results provide causal evidence for the role of theta frequency oscillations in the reactivation of a WM memory trace. Also, these results suggest an inverse relationship between theta and alpha frequency oscillations in reactivating and suppressing WM representations.

Disclosures: **J. Riddle:** None. **D. Cellier:** None. **S. Dhanani:** None. **M. Desposito:** None.

Poster

430. Neuromodulation and Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 430.03/VV4

Topic: H.02. Human Cognition and Behavior

Support: This work was supported by the national research foundation of Korea (NRF-2016R1A2B4010897)

This work was supported by the national research foundation of Korea (NRF-2016M3C7A1905475)

GIST Research Institute (GRI) grant funded by the GIST in 2017

Title: May mental arithmetic performance be enhanced by transcranial direct current stimulation? - Preliminary results

Authors: *S. JANG[1], *S. JANG[1], M. KWON[1], K. KIM[2,3], S. C. JUN[1]
[1]Sch. of Electrical Engin. and Computer Sci. (EECS), Gwangju Inst. of Sci. and Technol. (GIST), Gwangju, Korea, Republic of; [2]Ctr. for Biosignals, Korea Res. Inst. of Standards and Sci. (KRISS), Daejeon, Korea, Republic of; [3]Dept. of Med. Physics, Univ. of Sci. and Technol. (UST), Daejeon, Korea, Republic of

Abstract: Transcranial direct current stimulation (tDCS) is a non-invasive brain stimulation to modulate various cognitive functions such as working memory. The role of working memory in mental arithmetic has been studied in various research groups. Posterior parietal cortex (PPC) is known to be involved in arithmetic processing, but some researchers argued that dorsolateral prefrontal cortex (DLPFC) may also be involved in this processing. To verify this argument, we recorded magnetoencephalography (MEG) pre- and post-tDCS to find which area is strongly involved in multiplication problem. In this study, behavior tests were analyzed as a preliminary study.
Fifteen healthy volunteers (9 females, aged 24.5 ± 3.0 years) participated in our experiment. All subjects were given 3 stimulation types (anode, cathode, sham) on 3 other days (interval is at least one week). The current stimulation was delivered using a Starstim device (Neuroelectrics) via a pair of circular electrodes with sponge surface (25 cm^2). A 1.5 mA current intensity was applied for 25 min. The active electrode was placed over the left PPC at P3, and the reference electrode was located at Fp2 on the right DLPFC. Each participant performed mental multiplication problems and 152-channel whole-head MEG (KRISS) at a sampling rate of 1024 Hz was recorded.
Table 1 shows behavior results. A reaction time of solving the multiplication problems in post-tDCS was faster in anodal tDCS than those in cathodal and sham tDCS (anode vs. cathode:

p=0.028, anode vs. sham: p = 0.049), whereas there was no significant difference between cathodal and sham tDCS. Moreover, the reaction time (p = 0.078) and accuracy (p = 0.102) in only anodal tDCS were improved between pre- and post-tDCS. However, there was no significant difference in the reaction time and accuracy on the other stimulations.
The behavior results showed that anodal tDCS on the left angular gyrus associated with verbally mediated processing improves ability to solve the problems. It is known that the mental calculation is involved in phonological working memory related to arithmetic fact retrieval and procedural strategies. Thus, our preliminary results infer that activation in the left PPC may play some role of phonological working memory to solve the multiplication.

Table 1. Behavior analysis : Reaction time and accuracy of problem solving (Mean ± SD)

	Anode	Cathode	sham
Pre-tDCS			
Time (sec)	3.27 ± 1.50	3.29 ± 1.52	3.36 ± 1.56
Accuracy (%)	95.99 ± 2.35	95.33 ± 3.29	94.91 ± 4.28
Post-tDCS			
Time (sec)	3.13 ± 1.45	3.29 ± 1.54	3.31 ± 1.61
Accuracy (%)	97.22 ± 2.13	96.94 ± 2.27	96.94 ± 2.68

Disclosures: **S. Jang:** None. **M. Kwon:** None. **K. Kim:** None. **S.C. Jun:** None.

Poster

430. Neuromodulation and Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 430.04/VV5

Topic: H.02. Human Cognition and Behavior

Support: Air Force Office of Scientific Research Grant FA9550-10-1-0385

Title: Transcranial direct current stimulation during working memory training increases transfer to problem solving and alters connectivity between the dorsal attention network and trained cortex

Authors: *D. CISLER, M. STRENZIOK, A. HARWOOD, R. PARASURAMAN, P. GREENWOOD
George Mason Univ., Fairfax, VA

Abstract: An important goal of cognitive training is far transfer to fluid cognitive ability (Gf). Although working memory (WM) training has shown the strongest evidence to date of far transfer to fluid ability (e.g., Jaeggi et al., 2008), the literature is inconsistent. Previous work using transcranial direct current stimulation (tDCS) has shown benefits on n-back WM performance (meta-analysis of Brunoni & Vanderhasselt, 2014). Based on evidence that successful cognitive training modulated connectivity between nodes of the dorsal attention network (DAN) and trained cortex (Lewis et al., 2009; Strenziok et al., 2014), we hypothesized that cognitive training, in conjunction with anodal tDCS over the DAN, would facilitate far transfer and modulate functional connectivity involving the DAN.
Young adults were pre-tested and pre-scanned before being randomly assigned to adaptive WM training during either 2 mA or .1 mA (sham) tDCS to CP4 (cathode on left arm). The supervised training was 1.5 hours a day (with tDCS or sham for 30 min of that time) for 4 consecutive days on adaptive WM memory tasks, followed by post-testing and post-scanning. Pre- and post-testing included: spatial working memory, visuospatial attention, WAIS Letter-Number Sequencing, WAIS Matrix Reasoning, Wechsler Memory Scale, Logical Memory, and the Everyday Problems Test. Functional connectivity (fcMRI) images were acquired at rest with participants' eyes open in axial plane with a single-shot echo-planar sequence sensitive to change in the BOLD response (TR = 2500 ms, TE = 30 ms, 42 slices, $3mm^3$ resolution, flip angle 70°). A MANOVA on the behavioral data revealed effects of tDCS on transfer tasks (Wilks' lambda, p=.048). Only speed of spatial working memory and Everyday Problems Test (Willis & Marsiske, 1993) benefited from tDCS (p=.037). Analysis of the fcMRI data involved a whole-brain seed-based correlation analysis in the dorsal attention network (superior parietal cortex, SPC), using a 2 mm spherical region (MNI: -14, -69, 48) previously used to show changed fcMRI following sensory training (Lewis et al., 2009; Strenziok et al., 2014). Functional connectivity analyses demonstrated a Group x Time interaction ($p < .05$) in the lateral occipital lobe indicating a decrease in connectivity in the tDCS group and increase in the sham group, from pre-to post training. This extends to young adults and to WM our previous finding of perceptual training-related functional connectivity change in older adults (Strenziok et al., 2014). Effects of tDCS on changes in the fMRI BOLD signal as a function of size of cues around the memory array of a cued spatial WM task will be included.

Disclosures: D. Cisler: None. **M. Strenziok:** None. **A. Harwood:** None. **R. Parasuraman:** None. **P. Greenwood:** None.

Poster

430. Neuromodulation and Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 430.05/VV6

Topic: H.02. Human Cognition and Behavior

Support: NIMH Intramural Research Program

Title: Prefrontal high gamma contributes differentially to the multi-frequency MEG response to rest and working memory task activation

Authors: F. W. CARVER[1], D. Y. RUBINSTEIN[2], S. I. FRADKIN[1], T. HOLROYD[1], *R. COPPOLA[1]
[1]MEG Core Facility, [2]CTNB, NIMH, Bethesda, MD

Abstract: Human electrophysiological activity contains oscillations at several distinct frequency bands, the relative amplitudes of which can be modulated by factors such as alertness and the performance of tasks. In recent years, task-dependent signal change has been localized to specific brain regions utilizing high density EEG and MEG systems combined with advanced source localization techniques. Largely due to a low signal-to-noise ratio, the high end of the cortical power spectrum (~>60 Hz) has received relatively little experimental attention compared to higher amplitude bands such as theta (~3-8 Hz), alpha (~8-12 Hz), beta (~13-30 Hz), and gamma (~30-60 Hz). Here we employed a large sample of healthy volunteers (*n*=191) to characterize the oscillatory response to an n-back working memory task by contrasting the data to a resting eyes-closed recording using a 275-channel CTF system. Analysis of channel power spectra from 2-back, 0-back, and rest trials revealed signal reduction associated with greater task difficulty in alpha and beta bands, a well-known phenomenon known as event-related desynchronization. Significantly, the spectra also showed task-related increase in power in a broad band of high-frequency activity greater than 60 Hz, which researchers often refer to as high gamma band. Source localization of this activity using synthetic aperture magnetometry revealed a distinct distribution of sources for this high gamma activation compared to the alpha and beta bands. Activity in the band was especially prominent in prefrontal cortex, including Brodmann areas 9, 10, and 32, regions known to be critical for the executive functions such as attention and working memory necessary for the performance of the n-back task. These results indicate that the high gamma band is part of the neurophysiological instantiation of these, and other, cognitive processes. The data were collected as part of a larger study of the genetic contributions to the development of schizophrenia, a patient group known to exhibit abnormalities in prefrontal function. Our findings suggest that elucidating the high gamma band will be important for discovering the origin of these abnormalities, and their contribution to diminished cognitive function.

Disclosures: **F.W. Carver:** None. **D.Y. Rubinstein:** None. **S.I. Fradkin:** None. **T. Holroyd:** None. **R. Coppola:** None.

Poster

430. Neuromodulation and Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 430.06/VV7

Topic: H.02. Human Cognition and Behavior

Support: This study was supported by the German Federal Ministry of Education and Research (BMBF, grant 01GQ1102 to H.T.).

Title: Novelty modulates human striatal activation and prefrontal-striatal effective connectivity during working memory encoding

Authors: ***L. S. GEIGER**[1], A. SCHAEFER[1], C. MOESSNANG[1], Z. ZANG[1], M. ZANGL[1], H. CAO[1], T. VAN RAALTEN[2], A. MEYER-LINDENBERG[1], H. TOST[1]
[1]Central Inst. of Mental Hlth., University of Heidelberg, Mannheim, Germany; [2]Rudolf Magnus Brain Ctr., University Medical Center Utrecht, Utrecht, Netherlands

Abstract: The gating function of the basal ganglia (BG) in motor responses is well established and growing evidence supports an analogous role of BG during working memory (WM) encoding, a task phase in which gating of relevant materials is essential (e.g. McNab and Klingberg, 2008; Moore et al., 2013; O'Reilly and Frank, 2006). One important, but highly understudied aspect of stimulus relevance is the novelty of WM items. We therefore set out to study the effect of novelty on corticostriatal function and effective connectivity during an established WM task.
We used a modified Sternberg WM task (van Raalten et al., 2008) with different memory phases (encoding vs. retrieval) and degrees of stimulus familiarity (novel vs. previously practiced). FMRI data were acquired at 3T in a sample of 74 healthy participants (43 females, age: 26 ± 7.0 years). Data preprocessing was performed using standard routines of the Statistical Parametric Mapping software (SPM8; http://www.fil.ion.ucl.ac.uk/spm/software/spm8/). First level analyses included the calculation of individual contrast images to assess the effect of memory phase (encoding vs. retrieval) and effect of stimulus type (novel vs. practiced). Individual contrast images were subjected to a second-level random-effects model and one-sample t-tests were calculated for statistical inference at the group level ($P < 0.05$, whole-brain family-wise error corrected). Corticostriatal connectivity analyses were performed in two key regions, the dorsolateral prefrontal cortex (DLPFC) and the striatum using dynamic causal modeling (DCM) implemented in the DCM12 toolbox (SPM12). First, we defined model families based on different modulation patterns (2^4 = 16 families) and compared these families using Bayesian model selection. The models within the winning family (15 models) were subsequently

compared using random effects Bayesian model selection (BMS) to determine the winning model (Stephan et al., 2007).
Activation analyses demonstrated a significant engagement of the striatum and the DLPFC specifically during the encoding of novel WM items. DCM analyses identified a selective positive modulatory influence of novelty encoding on the connection from the DLPFC to the striatum. These data extend prior research by further underscoring the relevance of the BG for human cognitive function and provide a mechanistic account of the DLPFC as a top-down regulatory element of striatal function that may facilitate the gating of novel WM items. Further studies should address this gating mechanism in a longitudinal approach or in clinical populations with working memory dysfunction such as in patients with schizophrenia.

Disclosures: L.S. Geiger: None. **A. Schaefer:** None. **C. Moessnang:** None. **Z. Zang:** None. **M. Zangl:** None. **H. Cao:** None. **T. van Raalten:** None. **A. Meyer-Lindenberg:** F. Consulting Fees (e.g., advisory boards); A.M.-L. has received consultant fees from AstraZeneca, Elsevier, F. Hoffmann-La Roche, Gerson Lehrman Group, Lundbeck, Outcome Europe Sárl, Outcome Sciences, Roche Pharma, Servier International and Th. **H. Tost:** None.

Poster

430. Neuromodulation and Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 430.07/VV8

Topic: H.02. Human Cognition and Behavior

Support: SNSF 320030_156029

Title: Verbal working memory elicits synchronization between cortex and hippocampus

Authors: *J. SARNTHEIN[1], E. BORAN[1], P. HILFIKER[2], L. STIEGLITZ[1], T. GRUNWALD[2], T. FEDELE[1]
[1]Klinik für Neurochirurgie, Universitätsspital Zürich, Zurich, Switzerland; [2]Swiss Epilepsy Ctr., Zurich, Switzerland

Abstract: Objective: Verbal working memory elicits workload-dependent theta and alpha oscillations in the frontal and parietal surface EEG (Michels, 2008), but the involvement of subcortical nodes is not known.
Methods: Epilepsy patients with electrodes in the hippocampus and on the scalp performed a modified Sternberg task with setsize 4, 6 and 8 letters. We analyzed the time-frequency profile and the phase locking value (PLV) in theta (4-8 Hz) and alpha (8-12 Hz) and high gamma (> 100 Hz) frequency bands while stimuli were encoded and retained in memory.
Results: In 9 of 10 participants, the theta/alpha PLV was elevated between hippocampus and scalp. In 4 participants, the PLV increased with setsize, predominantly towards the end of the

retention period and to frontal and parietal electrodes. Two participants showed strong frontal midline theta and parietal alpha during retention, in agreement with our earlier scalp EEG study. Concurrently, the workload and/or the task conditions modulated the hippocampal theta/alpha and high-gamma power.
Conclusion: While hippocampal activity is known for visuospatial memory tasks, we show here hippocampal involvement also in a cortical network that is activated during verbal working memory and mediated by synchronized theta/alpha EEG oscillations.
Michels et al. (2008) EEG alpha distinguishes between cuneal and precuneal activation in working memory. Neuroimage 40(3):1296

Disclosures: J. Sarnthein: None. **E. Boran:** None. **P. Hilfiker:** None. **L. Stieglitz:** None. **T. Grunwald:** None. **T. Fedele:** None.

Poster

430. Neuromodulation and Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 430.08/VV9

Topic: H.02. Human Cognition and Behavior

Title: Exploring the effect of age, task performance and learning style on prefrontal hemodynamics during a working memory task

Authors: *A. A. ANDERSON[1], K. PARSA[1], S. GEIGER[2], R. ZARAGOZA[1], R. KERMANIAN[1], E. SMITH[1], A. GANDJBAKHCHE[1]
[1]NICHD, NIH, Bethesda, MD; [2]St. Olaf Col., Northfield, MN

Abstract: Existing literature outlines the quality and location of activation in the prefrontal cortex (PFC) during working memory (WM) tasks. However, the effects of individual differences on the underlying neural process of WM tasks are still unclear. In this functional near infrared spectroscopy study, we administered a visual and auditory n-back task to examine activation in the PFC while considering the influences of age, task performance, and preferred learning strategy (VARK score). Results showed that younger subjects had higher activation in the right ventro-medial-PFC compared to older subjects. Aside from age differences, high performance (HP) subjects (task accuracy > 90%) showed lower activation compared to normal performance subjects. After accounting for learning style we found a correlation between aural VARK score and level of activation in the PFC. Subjects with higher aural VARK scores displayed lower activation during auditory stimuli, while exhibiting higher activation during visual stimuli. High performance subjects during auditory task had higher aural and visual VARK scores, indicating an effect of learning style on the task performance and activation. The results of this study show that learning style and task performance can influence PFC activation,

with applications toward neurological implications of learning style and populations with deficits in auditory or visual processing.

Disclosures: **A.A. Anderson:** None. **K. Parsa:** None. **S. Geiger:** None. **R. Zaragoza:** None. **R. Kermanian:** None. **E. Smith:** None. **A. Gandjbakhche:** None.

Poster

430. Neuromodulation and Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 430.09/VV10

Topic: H.02. Human Cognition and Behavior

Support: NSF Grant BCS-1439188

Title: Higher working memory capacity is associated with better suppression of internal noise

Authors: *A. RAJAN, S. MEYYAPPAN, H. WALKER, M. DING
J. Crayton Pruitt Family of Dept. of Biomed. Engin., Univ. of Florida, Gainesville, FL

Abstract: Working memory capacity (WMC) is thought to measure an individual's ability to maintain executive attention to task relevant information in the presence of distraction. Past work testing this idea has mainly focused on external distraction in the form of environmental distractors. To what extent WMC also enables the suppression of internal distraction or noise, such as mind wandering mediated by the default mode network (DMN), is unknown. We addressed this problem by recording fMRI data from 20 subjects performing a cued attention task. Each trial started with an auditory cue, which directed subjects to attend to a spatial location (left or right visual field) or a color (red or green). Following a random time delay, two stimuli in the form of a colored rectangle were displayed in the right and left visual field. The subjects were required to discriminate the orientation (vertical or horizontal) of the rectangle in the cued location or cued color. The WMC of each subject was separately assessed by an automated operation span (OSPAN) task. We found the following results. First, posterior cingulate cortex (PCC) of the DMN was significantly deactivated in response to the cue, and the magnitude of deactivation was inversely correlated with the reaction time (RT) variability, namely, the stronger the PCC deactivation, the smaller the RT variability. Second, the magnitude of PCC deactivation was inversely correlated with WMC, namely, subjects with higher WMC had more deactivation in PCC in response to attentional cues. Third, a beta series connectivity analysis using PCC as the seed region revealed that PCC was positively correlated with the major DMN regions including medial prefrontal cortex, bilateral angular gyrus, bilateral parahippocampal region but anti-correlated with major task control regions including dorsal anterior cingulate, bilateral anterior insula, and bilateral dorsolateral prefrontal cortex. These results support the hypothesis that subjects with higher WMC are better at suppressing internal noise, leading to

reduced attention lapses, and such suppression is possibly implemented by the higher-order task-control networks such as the salience network and the central executive network.

Disclosures: **A. Rajan:** None. **S. Meyyappan:** None. **H. Walker:** None. **M. Ding:** None.

Poster

430. Neuromodulation and Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 430.10/VV11

Topic: H.02. Human Cognition and Behavior

Support: DoD Defense Health Program Grant NF90UG

Title: Examination of cerebral cortico-cortical communication for cognitive workload assessment during dual-task walking

Authors: ***E. P. SHAW**[1,2,3], J. C. RIETSCHEL[5], B. D. HENDERSHOT[1,6,7], A. L. PRUZINER[1,6,7], M. W. MILLER[8], B. D. HATFIELD[2,3], R. J. GENTILI[2,3,4]
[1]Rehabil., Walter Reed Natl. Military Med. Ctr., Bethesda, MD; [2]Neurosci. and Cognitive Sci., [3]Kinesiology, [4]Maryland Robotics Ctr., Univ. of Maryland, College Park, MD; [5]Veteran's Hlth. Admin., Baltimore, MD; [6]DOD-VA Extremity Trauma and Amputation Ctr. of Excellence, Bethesda, MD; [7]Rehabil. Med., Uniformed Services Univ. of the Hlth. Sci., Bethesda, MD; [8]Kinesiology, Auburn Univ., Auburn, AL

Abstract: While some prior work has employed ERPs and EEG spectral content to successfully evaluate cognitive workload during dual-task walking, none have examined changes in cerebral cortico-cortical communications; a critical component of brain dynamics during cognitive-motor performance. Therefore, this study aimed to examine whether changes in the magnitude of cortico-cortical communications could serve as an index of cognitive workload under various levels of cognitive challenge during dual-task walking. EEG was collected from twelve uninjured participants who were seated or walking on a treadmill in a Computer Assisted Rehabilitation Environment while performing a cognitive task of varying difficulty (easy or hard). Due to its common use in brain research, EEG coherence was employed to assess cortical networking. Additionally, the weighted phase lag index (WPLI) was computed as it is more robust to volume conduction. Both metrics were initially computed using a scalp montage centered over the motor planning region (i.e. electrode Fz). Such a montage was employed due to its common use in cognitive-motor studies. Statistical analysis of Repeated Measures ANOVAs revealed some significant contrasts of interest for both the difficulty of the cognitive task (easy/hard) and the condition performed (seat/walk) in EEG fronto-frontal coherence for the lower- to mid- frequency bands. However, these findings were not fully supported by the WPLI. Similar analyses of the mid- to high-frequency bands did not reveal differences in cognitive task

difficulty, but were moderately sensitive to differences in the performance conditions. Overall, the findings did not provide a clear assessment of cognitive workload in contrast to ERP and regional activation analyses employed in similar dual-task walking studies. Although dual-task walking has an important cognitive-motor component, it is possible that the montage considered here may not have been able to fully capture changes in cortical networking of uninjured individuals. This is because walking alone is likely to be more automatic (requires less motor planning) compared to upper-extremity task performance. Consequently, a more exhaustive analysis based on data mining is currently underway to capture more detailed patterns of cortical connectivity underlying various changes in task demands. This work was supported by the DoD Defense Health Program (NF90UG) and the DoD-VA Extremity Trauma & Amputation Center of Excellence. Views expressed are those of the authors and do not reflect the official policy or position of the Departments of the Army, Navy, or Defense, or the U.S. Government.

Disclosures: E.P. Shaw: None. **J.C. Rietschel:** None. **B.D. Hendershot:** None. **A.L. Pruziner:** None. **M.W. Miller:** None. **B.D. Hatfield:** None. **R.J. Gentili:** None.

Poster

430. Neuromodulation and Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 430.11/VV12

Topic: G.05. Anxiety Disorders

Support: NIH IRP ZIAMH002798

Title: Methylphenidate manipulation of the neural underpinnings of anxiety and working memory interactions

Authors: *B. FUCHS, T. LAGO, N. BALDERSTON, C. GRILLON, M. ERNST
Natl. Inst. of Mental Hlth., NIH, Bethesda, MD

Abstract: Anxiety can impair cognition, and cognitive processes can decrease anxiety. However, the neural mechanisms underlying these effects are unclear. Here, cognition and anxiety were manipulated orthogonally to examine their effects on the neural substrates of working memory (WM). Enhancing executive functioning pharmacologically with a single-dose of methylphenidate-MPH (vs. placebo-PLC) was expected to: (1) facilitate recruitment of the cognitive circuitry, and (2) free resources to attend to threat, i.e increased recruitment of the threat circuitry (vs. safe condition), particularly during high-load WM (vs. low-load).
In a placebo-controlled, double-blind, parallel-group fMRI study, 61 healthy volunteers received MPH (20 mg) or PLC, and performed 90min later an N-back WM task in the scanner (1- and 3-back), under both conditions of threat of shock or safety. Functional images were preprocessed and analyzed in AFNI. WM performance and BOLD activity were assessed using Drug (MPH,

PLC) x Threat (Safe, threat) x Load (1-back, 3-back) repeated measure ANOVAs. Behaviorally, in the MPH group, threat increased accuracy during 3-back and decreased accuracy in 1-back. In the PLC group, threat did not influence behavioral performance. Neurally, whole brain analyses revealed main effects of threat (insula, cingulate, inf parietal) and load (inf frontal, sup parietal, cingulate, med temporal). A threat x load ANOVA in the MPH group revealed an interaction in the superior parietal lobule (SPL), an area vital for WM. Post-hoc analyses in SPSS revealed a drug x threat x load interaction, where in MPH, threat increased SPL BOLD during the 3-back and decreased BOLD during the 1-back. This interaction was not seen in the PLC group.
Results suggest that MPH increased resources towards cognitive circuitry, resulting in improved accuracy, during threat in high load. The opposite effect of MPH on WM-related processing during 1-back suggests that the increased resource spills over to process task-irrelevant threat information, which interferes with the task at hand, resulting in decreased accuracy. However, MPH did not appear to modulate threat-related processing. Future work is necessary to further examine these anxiety and cognition interactions.

Disclosures: B. Fuchs: None. **T. Lago:** None. **N. Balderston:** None. **C. Grillon:** None. **M. Ernst:** None.

Poster

430. Neuromodulation and Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 430.12/VV13

Topic: G.05. Anxiety Disorders

Support: NIH Intramural ZIAMH00279

Title: The effect of threat of shock on working memory maintenance vs. manipulation during the Sternberg paradigm

Authors: *N. L. BALDERSTON, A. HSIUNG, V. ONYEACHU, M. ERNST, C. GRILLON
Natl. Inst. of Mental Hlth., NIH, Bethesda, MD

Abstract: Previous studies show that increasing working memory (WM) load reduces anxiety. One possible explanation for this effect is that anxiety and WM engage similar processing resources, and must compete for these resources when task demands are high. A key region implicated may be the right dorsolateral prefrontal cortex (dlPFC), which has been shown to play a role in both anxiety and WM. One way to test this hypothesis is to manipulate WM load using multiple methods (i.e. maintenance and manipulation), and examine whether these processes differentially engage the dlPFC when subjects are anxious. According to the shared resources hypothesis, anxiety should increase dlPFC activity similarly for maintenance and manipulation

processes.
Subjects performed a Sternberg WM task in an fMRI study. They were presented with a series of letters followed by a brief maintenance period. The task included 3 types of trials based on the number of letters presented and the instructions: maintain 5, maintain 8, and sort 5. On the maintain trials, subjects were instructed to maintain the letters in the order they were presented, while on the sort trials subjects were instructed to rearrange the letters in alphabetical order. These trials took place during blocks of safety and unpredictable shock threat.
BOLD responses were extracted to the trial types using a functional region of interest centered on the right dlPFC. We quantified maintenance activity by subtracting the BOLD responses to the maintain 5 trials from BOLD responses to the maintain 8 trials. We quantified manipulation activity by subtracting BOLD responses to the maintain 5 trials from BOLD responses to the sort 5 trials.
In healthy controls, maintenance-related dlPFC activity was increased by threat, whereas manipulation-related activity was elevated for both safe and threat blocks. Given that increasing WM load through maintenance but not manipulation leads to threat-related dlPFC increases, these results suggest that the shared resources hypothesis alone is not sufficient to explain the effect of WM load on anxiety. Instead, they support the hypothesis that increasing WM maintenance processes engages an active attention control mechanism when subjects are anxious, and that this attention control mechanism is also engaged by WM manipulation. Preliminary results in individuals diagnosed with generalized anxiety disorder suggest reduced dlPFC responses in general, with a similar pattern across the conditions, compared to controls.

Disclosures: **N.L. Balderston:** None. **A. Hsiung:** None. **V. Onyeachu:** None. **M. Ernst:** None. **C. Grillon:** None.

Poster

430. Neuromodulation and Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 430.13/VV14

Topic: H.02. Human Cognition and Behavior

Support: NIH Grant 5F31DC014230

Title: The impact of age of language exposure on spatial working memory using fNIRS neuroimaging

Authors: *G. KARTHEISER[1], L.-A. PETITTO[2]
[1]Brain and Language Lab. For Neuroimaging, Washington, DC; [2]Gallaudet Univ., Washington, DC

Abstract: Early-exposed (native) signers of signed languages have been shown to outperform hearing individuals with no signed language on behavioral tests of spatial working memory (e.g. Emmorey et al. 1993), presumably because of the spatial nature of signed languages. This permits a powerful test of hypotheses about the development of the neural circuitry underlying human spatial cognition and its relation to language. Spatial cognition is argued to be a capacity that is *not* vulnerable to sensitive periods in development, yet human language learning is widely understood to be highly impacted by the age of first language and second language exposure; early exposure to two languages produces positive, robust impact on neurocognitive development, even possibly prolonging the sensitive period of human language learning (Jasinska & Petitto, 2013). Using fNIRS to study adult first-time learners of a signed language (both spatial and a language), and varying proficiency in achievement (low and high), we ask whether the age of sign language exposure impacts the neural recruitment underlying spatial working memory and language? Age-effects were predicted to shed new light on sensitive periods for human spatial cognition.
Methods. Hearing signing adults, 3 Groups: Native Signers (N=5); Late adult learners of sign/Low Proficiency (N=5), and Late adult learners of sign/High Proficiency (N=5). Block design, 3 spatial n-back conditions: 0-back, 1-back, 2-back. Time-locked recorded fNIRS neuroimaging.
Results. fNIRS: n-back behavioral accuracy/speed scores were impacted by task difficulty, confirming task validity. As difficulty increased, Native Signers showed significant neural activation in L agranular frontal area ($p<.006$) and inferior frontal gyrus ($p<.006$). High Proficiency late adult sign learners showed significant activation in R agranular frontal area ($p<.006$); Low Proficiency showed significant activation in R frontopolar prefrontal cortex ($p<.023$).
Preliminary findings suggest that spatial working memory is sensitive to *age* of exposure. Human signed languages permit a new way to examine the malleability of other higher cognitive functions—here, spatial cognition—and permit insights into the brain's structural and functional plasticity. The findings also advance scientific debate about the nature of the flexibility and reversibility of sensitive periods in adult learning. This work has potential for high translational significance as it suggests early sign language exposure may be used as a novel tool in building spatial cognition for all individuals, as it has been shown that spatial abilities greatly predict STEM success.

Disclosures: G. Kartheiser: None. **L. Petitto:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.01/VV15

Topic: H.02. Human Cognition and Behavior

Title: Memory in autism: a preliminary fmri study

Authors: *S. HUEMER
Psychology, Loyola Marymount Univ., Los Angeles, CA

Abstract: Background: The Remember/Know (R/K) procedure (Tulving, 1985, 2002) is used in recognition tasks to study both semantic and episodic memory systems. Zelazo and Frye (2001) have shown that children with ASD have problems with episodic remembering. Episodic recognition involving the recollection of contextual information (R) is mediated by hippocampal processes while familiarity based recognition (K), which is intact in ASD, is mediated by perirhinal processes (Brown & Aggleton, 2001). Morphological abnormalities of the hippocampus are well documented in ASD (Groen et al., 2010; see Nicolson et al., 2006). Objectives: Our study was interested in finding out how adolescents with ASD process their own first name as opposed to other names and compared to neurotypical peers. We predicted that control subjects would activate areas of self-reference and episodic memory, such as the left tempo-parietal cortex, the superior temporal gyrus, and the hippocampus when hearing their own name. Subjects with ASD, on the other hand, were expected to employ compensatory processes including the perirhinal regions when hearing their own name. Methods: In this preliminary functional magnetic resonance imaging (fMRI) investigation, we compared brain activity of 9 adolescents with ASD with those of 9 neurotypical controls to four categories of auditory language stimuli: their own first name, familiar people's names, names of objects of high interest, and numbers. Each participant listened passively to the randomly sequenced names five times during three sessions. We administered a test of verbal receptive ability to test whether our results may align with scores on the test. Results: When hearing their own names, controls and subjects with ASD who scored high on the verbal ability test activated regions in the insula, superior temporal gyrus as well as the hippocampus, brain regions associated with self-referential processing and long term memory, or, R (remembering). In contrast, subject with ASD who scored low on our test of receptive verbal ability lower-scoring subjects showed activity in the prefrontal cortex and the thalamus, regions associated with new learning and K (knowing). Conclusion: Our findings suggest that ASD subjects with lower verbal ability recognize, or, "know" their own name like a newly acquired fact rather than "remember" their name in a self-referential and spatio-temporal context. The results of controls and ASD subjects with higher verbal ability were more like each other than the results of the higher and the lower scoring ASD groups, which indicates that memory function in ASD aligns with verbal ability.

Disclosures: S. Huemer: None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.02/VV16

Topic: H.02. Human Cognition and Behavior

Support: NSC 103-2420-H-002 -009 -MY3

Title: Intervention of memory consolidation by post-trial learning: An fMRI study on figure-word associative memory

Authors: *T. CHOU[1], F.-W. WU[1], K.-C. LIANG[2]
[2]Psychology, [1]Natl. Taiwan Univ., Taipei, Taiwan

Abstract: Memory consolidation is an off-line process in which a memory trace is growing and gradually resisting to interference. It involves changes of neural substrates requiring hippocampal-cortical interaction. Interruption of this process, such as introducing a post-encoding task, reduces memory strength and hence the retrieval performance in a test. To address the issue, this study probed the effect of a post-encoding task on memory consolidation of pair association. By using task-related fMRI, we observed the brain activity during periods of retrieval and consolidation in particular. The experiment contained a learning phase and a retrieval phase that were separated by 24 hrs. Thirty-eight participants were instructed to learn two sets of lists containing 48 pairs of fractal figures with words: a target list and an intervention one. After learning the target list, participants learned the intervention list within an hour. The memory performance was examined both immediately (day 1) and 24 hrs (day 2) after learning by recognition. The results showed that recognition of the figure-word association was accompanied with greater activation in the hippocampus, anterior cingulate cortex, and posterior cingulate cortex for day 1 as compared to day 2. In contrast, recognition of the figure-word association was accompanied with greater activation in the left inferior frontal gyrus and right occipital cortex for day 2 as compared to day 1. Greater inferior frontal activation is thought to reflect increased demands on retrieving verbal information of figure-word associative memory. Moreover, greater occipital activation is associated with analyzing visual information of figure-word associative memory. These findings suggest that a post-encoding task may affect memory formation and hippocampal-cortical connectivity, implying that consolidating figure-word associative memory in humans may require reverberating activity among the hippocampus, inferior frontal gyrus, and right occipital cortex.

Disclosures: **T. Chou:** None. **F. Wu:** None. **K. Liang:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.03/VV17

Topic: H.02. Human Cognition and Behavior

Support: NIH Grant NS089729

Title: Spatiotemporal neural activity dissociates encoding and retrieval states

Authors: *N. M. LONG, M. L. DRASCHER, B. A. KUHL
Univ. of Oregon, Eugene, OR

Abstract: When you reencounter someone whom you've previously met, you may find yourself trying to retrieve past details about them only to realize that you have been ignoring what they are presently saying to you. This dichotomy between encoding and retrieval states has been demonstrated in rodents (Hasselmo et al., 2002) and recently in humans using fMRI (Richter et al., 2016). Here we conducted a human scalp electrophysiological study using multi-variate pattern analysis to test for dissociable representations of encoding and retrieval states. We employed a task in which participants viewed two lists of object pictures, with pairs of similar objects distributed across lists. For example, list 1 might contain a picture of an apple and list 2 would contain a picture of a different apple. Critically, during the second list participants were cued, on each trial, to either ‘retrieve’ (think back to) the item from list 1 (A) or to ‘encode’ the newly presented item (A'). Notably, even when the instruction was to retrieve, participants still had to process the second list item (A') in order to know which first-list item (A) they were to retrieve. Thus, in both conditions, participants were required to process current sensory information. Following several blocks of these paired lists, participants completed a forced-choice recognition post-test. On each trial of the post-test, participants were shown two similar objects (e.g., two apples) and were instructed to select the object they had previously seen. Memory for all list 1 (A) and list 2 (A’) objects was tested, but list 1 and list 2 items were tested on separate trials. For example, the apple from list 1 would be paired with a novel apple and the apple from list 2 would be paired with an additional novel apple. Behaviorally, instructions during list 2 influenced memory on the post-test. ‘Retrieve’ and ‘encode’ instructions were associated with relatively better memory for list 1 and list 2 items, respectively. This behavioral evidence indicates that subjects were able to modulate internal memory states. Pattern classification analyses revealed that encoding and retrieval states were also associated with dissociable patterns of spectral EEG activity. In part, these states were associated with shifts in high frequency activity. Collectively, these findings indicate that memory is influenced by shifts in processing states and that processing states are associated with distinct electrophysiological signals.

Disclosures: **N.M. Long:** None. **M.L. Drascher:** None. **B.A. Kuhl:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.04/VV18

Topic: H.02. Human Cognition and Behavior

Support: NIH Grant R01MH074692

NIH Grant F32MH106266

Title: Disentangling interactions between context switches and repetition effects

Authors: *L. J. LOHNAS, L. DAVACHI
New York Univ., New York, NY

Abstract: Improved memory for spaced over massed repetitions is a well-established finding (Cepeda, Pashler, Wixted and Rohr, 2006), and more recently it has been shown that a retrieved context model can account for these effects (Lohnas Siegel & Kahana, 2014). Further, encoding contexts have been shown to modulate repetition effects: whereas recall probability is greater for massed items repeated in different contexts, recall probability is greater for spaced items repeated in the same context (e.g., Verkoeijen, Rikers & Schmidt, 2005; Maskarinec & Thompson, 1976). Here we consider the role of context switches, as such switches modulate memory and attentional processes (Ezzyat & Davachi, 2014; DuBrow & Davachi, 2013) yet have not been considered in the aforementioned result. Twenty participants were presented with twelve lists of words while undergoing functional magnetic resonance imaging (fMRI). Participants performed free recall and source recognition for each list. During encoding, each word had an associated task context, and could be presented once, twice massed (lag=0) or twice spaced (lag=5). At each lag, words could be repeated with the same or different encoding contexts. For a massed item presented with different contexts, the second presentation always immediately follows a context switch, but the improved memory for these items has not yet been considered with respect to repetition effects. We thus introduced a matched control to the massed items, such that half of the spaced items repeated in different contexts had the second presentation immediately following a context switch, and half had the second presentation two items after the context switch. Using univariate and multivariate approaches, our results reflect how switches in encoding contexts modulate fMRI activity as well as successful encoding of repeated items.

Disclosures: L.J. Lohnas: None. **L. Davachi:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.05/VV19

Topic: H.02. Human Cognition and Behavior

Support: NIH Grant 1P01HD080679

Leverhulme Trust grant RPG-2014-075

Wellcome Trust Senior Investigator Award WT106931MA

Title: Conceptual knowledge modulates visual representations through a frontoparietal network

Authors: *K. BRAUNLICH[1], B. C. LOVE[1,2]
[1]Univ. Col. London, London, United Kingdom; [2]The Alan Turing Inst., London, United Kingdom

Abstract: Concepts can emphasize certain aspects or features of stimuli through selective attention. For example, the size of garments is critical when choosing what to purchase whereas weight may be more critical when choosing how to ship them. In an fMRI study, participants learned about two contrasting concepts, consisting of visual stimuli varying along four binary dimensions (position, size, shape, and color), through trial-and-error learning. A cognitive model of categorization was fit to individuals' categorization decisions. These model fits provided an attention weight for each of the four stimulus dimensions for each participant. Focusing on occipitotemporal cortex, we found that the more a stimulus dimension was attended, the better we could decode what value (e.g., red or blue) the dimension displayed on each trial. These results indicate a top-down influence of conceptual knowledge on visual representations of objects with this influence well characterized by a successful cognitive model of human categorization. To better understand the neural bases of these top-down concept-driven attentional effects, we investigated how these multivariate occipitotemporal representations interacted with activity elsewhere in the brain. To do so, we projected the occipitotemporal neuroimaging data onto a lower-dimensional subspace, which was spanned by a set of basis vectors that provided an estimate of both the sign (i.e., the evidence towards one particular dimensional attribute over the other, e.g., red vs. blue) and the strength of the multivariate occipitotemporal representation for each distinct perceptual dimension. We then employed a full-brain connectivity analysis to identify regions that might interact with these estimates of occipitotemporal representation. Activity in the right frontal eye fields, right inferior frontal sulcus (IFS), and bilateral intraparietal sulcus was consistent with a role in reading out information about the individual dimensional attributes. IFS, which is thought to be a part of a domain-general network associated with executive control, also carried general information (not tied to a specific feature value) about which stimulus dimensions were critical to the categorization decision, as well as signaled the overall entropy of the category decision. These results suggest that IFS plays an important role in modulating attention to specific perceptual dimensions by integrating knowledge of category structure, perceptual uncertainty, and overall decisional confidence.

Disclosures: K. Braunlich: None. **B.C. Love:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.06/VV20

Topic: H.02. Human Cognition and Behavior

Support: NIH Grant 1P01HD080679

Leverhulme Trust grant RPG-2014-075

Wellcome Trust Senior Investigator Award WT106931MA

Title: Estimating the functional dimensionality of neural representations

Authors: *C. AHLHEIM[1], B. C. LOVE[2,3]
[1]Exptl. Psychology, [2]Univ. Col. London, London, United Kingdom; [3]The Alan Turing Inst., London, United Kingdom

Abstract: Recent advances in multivariate fMRI analysis make it possible to distinguish fine-scaled differences in evoked brain patterns. Key to interpreting these patterns is estimating the underlying dimensionality of neural representations. Dimensions may correspond to psychological dimensions, such as length and orientation, or involve other coding schemes. Unfortunately, when adopting a naïve analysis approach, the noise structure of fMRI data inflates dimensionality estimates to the extent that data with no underlying dimensionality can display maximal dimensionality. To address these and related issues, we developed a novel approach that identifies brain regions that carry reliable task-modulated signal and estimates the signal's functional dimensionality.
Data takes the form of a voxel x condition (e.g., stimulus, task, etc.) matrix of beta estimates, resulting from a general linear model estimation of the BOLD response. Our approach uses a low-rank approximation technique, singular value decomposition (SVD), coupled with nested cross-validation to find the best low-dimensional projection of a pattern of voxel-responses on a single-subject level. Quality of the low-dimensional projections is measured as Pearson correlation between the projected and a held-out test set. To establish the validity of the technique, we conducted model recovery studies with simulated (i.e., synthetic) fMRI data. These checks indicated that our approach robustly estimates the underlying functional dimensionality across varying noise levels. With the soundness of the method established, we applied it to three published fMRI data sets involving conceptual and visual object recognition tasks. For the first concept learning dataset, functional dimensionality was found in occipital and fronto-parietal areas that heavily overlapped with the bespoke model-based fMRI analysis results reported in the original study, which bolstered the authors' original interpretation of their results. In a second concept learning fMRI dataset, we found that concepts that involved integrating

more stimulus features engendered higher-dimensional representations in a lateral occipito-temporal region of interest, which again elaborated the authors' original aims. Finally, we discuss results from a third study dissociating object shape and category. Together, these results indicate the theoretical value of identifying and characterizing the dimensionality of neural representations. We argue that doing is a necessary precursor to evaluating specific theories of neural function as the presence (or absence) of dimensionality constrains the interpretation of data.

Disclosures: C. Ahlheim: None. **B.C. Love:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.07/VV21

Topic: H.02. Human Cognition and Behavior

Support: NIH Grant 1P01HD080679

Leverhulme Trust Grant RPG-2014-075

Wellcome Trust Senior Investigator Award WT106931MA

Title: Measures of neural similarity

Authors: *S. BOBADILLA SUAREZ, B. C. LOVE
Exptl. Psychology (UCL), Univ. Col. London, London, United Kingdom

Abstract: One central goal in cognitive neuroscience is to understand how the brain represents information. In short, what makes two brain states similar? For example, when viewing a sparrow, representations in the visual system should be more similar to those of a robin than to those of a truck. When using fMRI to measure brain states, it is commonly assumed that Pearson correlation across voxels is an appropriate measure of neural similarity (i.e., when the voxel representations elicited by two stimuli correlate, then the brain states are similar), but this major theoretical assumption has not been fully evaluated. For example, Pearson correlation assumes that overall levels of (task-related) voxel activity are normalized and that each voxel independently contributes to similarity. Whereas, for instance, Minkowski measures assume similarity involves distances in a metrical space instead of vector directions or angles between vectors. Relatedly, the Mahalanobis measure expands on both measures by assuming the distributional pattern of voxel activity is consequential. Which similarity measure best describes the brain's operation? In this project, we endeavour to answer this question by evaluating a range of possibilities, including Pearson correlation. We address open questions such as whether the nature of neural similarity is common across tasks and brain regions. To that end, we follow the

tradition of grounding similarity in confusability; when two things are similar they are easily confused. Confusability of different brain states can be measured with classification procedures such as linear support vector machines (SVM). For example, a sparrow may be more likely to be misclassified as a robin than a truck. We then consider which similarity measure (from a large set) best characterizes this confusability data for each brain region and task considered. Across two previously published fMRI datasets, we find that similarity measures perform comparably across brain regions within a task, but that the best similarity measure is strongly task-dependent. In particular, a visual categorization task that involves stimuli that readily decomposes into parts is most consistent with Minkowski measures whereas a stimulus set involving natural images is most consistent with the Mahalanobis measure of similarity. We discuss possible causes for these task-specific neural representations and implications for multivariate analyses of neural data.

Disclosures: S. Bobadilla Suarez: None. **B.C. Love:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.08/VV22

Topic: H.02. Human Cognition and Behavior

Support: NSF BRAIN grant BCS-1533511

Title: Detecting content-specific patterns using targeted memory reactivation

Authors: B. WANG[1], J. W. ANTONY[1], S. LURIE[1], K. A. PALLER[2], *K. NORMAN[1]
[1]Princeton Univ., Princeton, NJ; [2]Dept. of Psychology, Northwestern Univ., Evanston, IL

Abstract: The replay of learning-related hippocampal and neocortical activity patterns is thought to drive memory stabilization. In this study, we attempted to decode replay in the electroencephalogram (EEG) in order to predict the consolidation of specific, recently-learned information. Human participants first encoded objects in one of four locations on the left or right part of the screen. Each object was accompanied by an inherently related sound (e.g. “moo” for cow) that we used to target memories during sleep. During encoding, we instructed participants to imagine moving their corresponding hand (i.e., left hand for objects on the left side, right hand for objects on the right side) towards the object for five seconds, after which they were instructed to execute the movement, touching the object picture on a touch-sensitive screen. As expected, participants showed significant lateralized activity for left- and right-hand trials during the imagination period. We then tested participants on the location of each object before they took an afternoon nap. During online indications of slow-wave sleep (SWS), we presented half of the sound cues associated with left- and right-hand movements. Finally, after a 2.5 hr break, participants returned for a final memory test. Behaviorally, participants remembered the correct

hemifield (left vs. right) significantly better for cued than uncued items. Neurally, we found lateralized EEG activity elicited by cues during sleep that discriminated between sounds linked (during wake) with left- vs. right-hand movements, thereby signaling the reactivation of learning-related memory content. Further analyses will determine the extent to which these asymmetries predict subsequent memory.

Disclosures: B. Wang: None. **J.W. Antony:** None. **S. Lurie:** None. **K.A. Paller:** None. **K. Norman:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.09/VV23

Topic: H.02. Human Cognition and Behavior

Support: NIH Grant R00MH103401

Title: Effects of contextual reinstatement on retrieval of item-emotion associations

Authors: *R. SAMIDE, K. KURKELA, M. RITCHEY
Dept. of Psychology, Boston Col., Chestnut Hill, MA

Abstract: Reinstating the encoding context during retrieval has been shown to enhance item recognition and recall, whereas switching contexts between encoding and retrieval may hinder memory performance. However, it is unknown whether contextual reinstatement confers memory benefits to non-contextual associations learned in that context, such as the emotional significance of an item. To fill this gap in the literature, we tested the effects of contextual reinstatement on memory for items and item-emotion associations. Participants viewed short blocks of neutral object images overlaid on one of four visual contexts, and for each block they were cued to learn that the objects were either ‘bombs’ or ‘safe.’ The bomb/safe designation was balanced across contexts, such that valence and context were orthogonal to one another. To imbue the ‘bomb’ stimuli with emotional salience, they were accompanied by an unpredictable startle probe (250ms burst of white noise) on 25% of trials. Immediately after encoding, participants completed a retrieval phase that assessed item recognition and emotional association memory. Memory sensitivity for the item-emotion association was assessed using a modified d’ measure (‘emotion discriminability index’). Importantly, during retrieval, items were shown overlaid on either the same context as encoding or a different context, which served as our manipulation of contextual reinstatement. Unexpectedly, emotion discriminability was significantly greater when the retrieval context was different from the context presented at encoding, indicating that under some conditions, contextual reinstatement can actually disrupt memory for non-contextual associations. Future research using fMRI will examine brain activity

related to the retrieval of emotional information in the presence or absence of contextual information.

Disclosures: **R. Samide:** None. **K. Kurkela:** None. **M. Ritchey:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.10/VV24

Topic: H.02. Human Cognition and Behavior

Support: Basque Government PhD fellowship from Department of Education, University and Research, PRE_2016_2_0116

MINECO Grant RYC-2014-1544

MINECO Grant PSI2015-65696

MINECO Grant SEV-2015-049

Basque Govenment (Dpt. Educacion, politica linguistica y cultura) Grant PI2016-12

Title: Testing leads to consolidated-like memories

Authors: *J. ARNAEZ-TELLERIA[1], M. CARREIRAS[1,2], P. M. PAZ-ALONSO[1]
[1]Basque Ctr. On Cognition Brain and Language, Donostia-San Sebastian, Spain;
[2]IKERBASQUE, Basque Fndn. for Sci., Bilbao, Spain

Abstract: Testing during encoding facilitates memory consolidation relative to classical repeated-study. This facilitation is known as the testing effect, which constitutes a robust phenomenon widely replicated. The retention interval (RI) between encoding and the final retrieval test seems to play a key role in this effect, with RIs longer than a day showing stronger results compared to RIs of a few hours. This finding points out the potential critical role of consolidation processes in the testing effect. As critical as it is sleep for memory consolidation, the testing effect has not been studied manipulating sleep periods. The present MRI study was aimed at investigating the functional and structural correlates underlying the effects of sleep in the retrieval of episodic memories encoded via repeated retrieval (RR) or repeated study (RS). To this end, 80 young adults divided in 4 groups participated. Participants encoded 180 Swahili-Spanish word pairs (e.g., *rafiki-amigo*) either under repeated retrieval or repeated study conditions. Participants came back three times and performed cued-recall tasks on 60 studied and 20 new non-studied items at 12 hours, 24 hours and 7 days after the initial encoding. Critically, participants were divided in 4 groups based on the initial encoding procedure (RT, RS) and

having or not an immediate sleep period after the initial encoding (Sleep, Wake): Repeated Retrieval Sleep (RRS), Repeated Retrieval Wake (RRW), Repeated Study Sleep (RSS) and Repeated Study Wake (RSW).
Behavioral results showed the beneficial effect of RR relative to RS across the three RIs, with one exception: the RSS group exhibited a similar performance to the RRS and RRW in the shortest RI (12 hours). However, this beneficial effect of sleep for the RSS group vanished in the longer RIs (24 hours and 7 days). For the RS groups, neuroimaging data revealed stronger hippocampal engagement for sleep (RSS) versus wake (RSW) groups during successful memory retrieval in the shortest RI (12 hours). Interestingly this difference also vanished in the longer RIs, with both RSS and RSW showing similar hippocampal engagement during successful memory retrieval after 24 hours and 7 days RIs. Functional connectivity analyses revealed tighter coupling among distributed hippocampal-PFC regions for successful retrieval of items studied under RR versus RS conditions in all three RIs and independently of the sleep/wake condition. Altogether, behavioral and neuroimaging data suggest that information studied via RR formed consolidated-like memories, while memories created via RS were more susceptible to benefit from regular consolidation processes.

Disclosures: J. Arnaez-Telleria: None. **M. Carreiras:** None. **P.M. Paz-Alonso:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.11/VV25

Topic: H.02. Human Cognition and Behavior

Support: NIH Intramural Research Program

Title: Dynamic changes in directed connectivity during paired associates memory task

Authors: R. YAFFE[1], J. I. CHAPETON[2], S. K. INATI[1], *K. A. ZAGHLOUL[3]
[1]Natl. Inst. of Neurolog. Disorders and Stroke, Bethesda, MD; [2]NINDS, NIH, Bethesda, MD; [3]Surgical Neurol. Br., Natl. Inst. of Neurolog. Disorders and Stroke, NIH, Bethesda, MD

Abstract: We investigated functional connections in human intracranial EEG data during paired associates memory encoding and cued recall. Our approach capitalizes on an important feature of functional connections that mediate communication in the human brain. Whether through direct or indirect synaptic connections, changes in local field potential signals coordinated across regions, or even the propagation of electric fields, communication between brain regions must occur through a physical connection of some kind. Importantly, the constraints associated with any physical connection imply that the time required for communication should be well defined and preserved. Every time information is conveyed from one point to another, it should take

approximately the same amount of time to do so. Hence, one plausible requirement for identifying a functional connection between brain regions is that the communication between them occurs with a consistent time delay. At any given moment in time, however, the amount of information being transferred may vary.
Here, we investigate functional connections in iEEG recordings as patients with medically refractory epilepsy participate in a paired-associates verbal memory task. Using the temporal precision afforded by iEEG, we used time-lagged mutual information (MI) to identify functional connections in the human brain that exhibit a consistent and significant increase in MI with a specific time delay. Mutual information (MI) is attractive in that it captures all temporal relations between two brain regions, and is agnostic to the particular neural mechanism underlying how those regions are communicating. We identify connections that have a consistent latency and change their level of communication dynamically in response to the presentation of memory stimuli. Our results suggest that we can identify connections in the brain that increase or decrease their flow of information during memory encoding and retrieval.

Disclosures: R. Yaffe: None. **J.I. Chapeton:** None. **S.K. Inati:** None. **K.A. Zaghloul:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.12/VV26

Topic: H.02. Human Cognition and Behavior

Support: SCHW1357/14-1

Title: Neural signature of successful memory updating

Authors: *L. M. KLUEN[1], G. JOCHAM[2], L. SCHWABE[1]
[1]Cognitive Psychology, Univ. of Hamburg, Hamburg, Germany; [2]Ctr. For Behavioral Brain Sciences, Otto-von-Guericke-University Magdeburg, Magdeburg, Germany

Abstract: In everyday life, we often experience situations in which existing memories turn out to be incorrect, requiring their modification in the light of new information. Although several lines of recent research indicate that memories are highly dynamic entities, how exactly established memories can be updated is largely unknown. In order to unravel the neural processes involved in successful updating of existing memories, we tested 48 participants in a novel memory updating paradigm on three consecutive days. On day 1, participants memorized face-city associations followed by a cued recall test for these associations. On day 2, participants saw the face stimuli again, while fMRI was recorded. Part of the faces were paired with the same city names they had been associated with the day before, while others were now presented with novel city names, requiring participants to update their memories. On day 3, participants

completed a recognition test, to assess whether the updating of their initial memories was successful. Results indicated that participants learned the initial face-city associations very well. Memory performance in the recognition test was generally high but memory updating was successful for only about 35 percent of the stimulus pairs that were altered on day 2. Our neuroimaging data revealed that successful memory updating, a key feature of adaptive memory, was linked to the recruitment of the dorsolateral prefrontal cortex during the presentation of information conflicting with established memories.

Disclosures: L.M. Kluen: None. **G. Jocham:** None. **L. Schwabe:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.13/VV27

Topic: H.02. Human Cognition and Behavior

Support: 973 Program (2014CB846102)

Title: Dopamine gene polymorphism, prefrontal cortex activation and neural pattern similarity during episodic memory encoding

Authors: *L. ZHENG, G. XUE
State Key Lab. of Cognitive Neurosci. and Learning, Beijing, China

Abstract: Neural pattern similarity across multiple learning events, which is posited to reflect the fidelity of cortical representation of the studied materials, is an effective indicator of subsequent memory performance. Although the role of dopamine in enhancing the fidelity of cortical representation has been implicated in computational and animal studies, evidence from human subjects is still rare. Using fMRI and a face-name association task, we investigated the relationship between neural pattern similarity and subsequent memory performance in more than 400 subjects. We further examined the neural and genetic factors that contributed to the individual differences in neural pattern similarity. We found that neural pattern similarity in the ventral visual cortex could predict the subsequent associative memory performance across items and individual subjects, even after controlling the activation level in these regions. Meanwhile, the prefrontal cortex activation level was also associated with better memory performance across items and individual subjects. Importantly, we found the prefrontal cortex activation was associated with greater neural pattern similarity in the VVC, which were in turn associated with better memory performance. Furthermore, the prefrontal cortex activation level was associated with individuals' dopamine beta hydroxylase (DBH) gene polymorphisms. Our results suggest a critical role of dopamine neurotransmitters and prefrontal cortex in enhancing neural pattern similarity and episodic memory encoding.

Disclosures: **L. Zheng:** None. **G. Xue:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.14/VV28

Topic: H.02. Human Cognition and Behavior

Title: Context-dependent memory effects revealed by reinstated neural oscillations

Authors: ***M. J. WÄLTI**, D. G. WOOLLEY, N. WENDEROTH
Neural Control of Movement Lab., ETH Zurich, Zuerich, Switzerland

Abstract: Many theories of memory rely on a mechanism called context-dependent memory, which refers to the finding that learnt information can be recalled better when the context of the test setting matches the encoding setting. A widespread method for investigating context-dependent memory effects has been the manipulation of simple context features, for example on a computer screen.
Here we performed a series of behavioral experiments to investigate whether context-dependent memory effects differ when encoding occurs in increasingly enriched visual environments. Based on an a-priori power analysis, each experiment included 40 participants who encoded 24 words embedded in different visual contexts. Subsequently, the words were freely recalled while the encoding context was either reinstated or not.
Experiment 1 was a replication experiment of Isarida and Isarida (2007), where we investigated the effect of reinstated background colors on free recall memory performance. We found no significant difference between reinstated versus not reinstated recall ($t(39) = -1.309$, $p = 0.198$).
Experiment 2 investigated the effect of using two distinct landscape images as backgrounds on a computer screen. However, the context again had no significant influence on recall ($t(39) = -1.723$, $p = 0.093$).
Experiment 3 provided two full-surrounding virtual reality environments during encoding. Again, no significant context effect on recall ($t(39) = 0.934$, $p = 0.356$) was observed, even in this highly immersive visual environment.
Finally, in **Experiment 4** we examined whether visual stimuli flickering at different frequencies that can potentially reinstate neural activity via sensory entrainment during encoding and retrieval would influence memory performance. Subjects observed flickering stimuli in the background on a computer screen to induce steady-state visually evoked potentials at 6 or 15 Hz in visual areas of the brain. We found that reinstatement of specific steady-state visually evoked potentials significantly improved recall performance compared to not reinstated conditions ($t(39) = 2.097$, $p = 0.043$).
The presented results indicate that effects of visual contexts on recall memory performance are

rather small, potentially because they are overshadowed by individual mnemonic strategies. We suggest that in Experiment 4, the flickering background and induced oscillations in visual areas are highly salient, i.e. more difficult to be ignored by the participant, and are therefore more likely to have an influence on the memorization/recall process.

Disclosures: M.J. Wälti: None. **D.G. Woolley:** None. **N. Wenderoth:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.15/VV29

Topic: H.02. Human Cognition and Behavior

Support: NIH HD67359

Title: Cortical oscillations during a fact-based memory integration task relate to academic success

Authors: *N. L. VARGA, J. R. MANNS
Psychology, Emory Univ., Atlanta, GA

Abstract: The acquisition and flexible extension of knowledge depends on the capacity to integrate across overlapping experiences. Memory integration refers to the process by which separate yet related memories are combined to support the derivation of new information. Recent functional magnetic resonance imaging (fMRI) studies have indicated that interactions between several cortical areas support memory integration in adults. Yet an open question concerns how the millisecond-level dynamical interactions between cortical regions involved in memory integration unfold in real-time. Thus, the goal of the present study was to investigate the rapidly-unfolding processes of memory integration by measuring oscillatory power and synchrony in scalp-based electrophysiological (EEG) recordings. Adults read novel, related facts (e.g., *Apple seeds are call pips; Cyanide is found in pips*) and were tested for self-derivation of novel integration facts (*Apple seeds contain cyanide*) while EEG was recorded. Alpha oscillations were prominent in the EEG and were observed to show marked second-by-second changes during encoding of the original, directly learned facts. Moreover, analyses of alpha coherence between clusters of EEG channels revealed that cortical alpha synchrony was greatest in a left-parietal cluster during encoding of the second, related fact (i.e., the first opportunity for memory integration) as compared to the first fact. This integration-related alpha synchrony was most pronounced in participants who subsequently showed enhanced memory integration performance. Integration-related alpha synchrony also showed marked differences between high and low performing participants when split by measures of scholastic achievement (i.e., college grade point average and SAT scores). Together, the findings inform our understanding of the

dynamical interactions between cortical regions supporting this fundamental learning ability, and have implications for how these processes relate to real-world academic success.

Disclosures: **N.L. Varga:** None. **J.R. Manns:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.16/VV30

Topic: H.02. Human Cognition and Behavior

Title: Illusory shape processing during encoding of scrambled items induces subsequent false memory for intact shapes

Authors: *J. M. KARANIAN[1,3], S. D. SLOTNICK[2]
[1]Psychology, [2]Boston Col., Chestnut Hill, MA; [3]Psychology, Wesleyan Univ., Middletown, CT

Abstract: In a recent study, we assessed whether the lateral occipital complex (LOC) – a conscious shape processing region – was associated with false memory for intact shapes (Karanian & Slotnick, under review). In that study, participants viewed intact or scrambled colored abstract shapes during encoding. During retrieval, colored disks were presented and participants indicated whether the corresponding item was previously "intact" or "scrambled". Like true memory for intact shapes, we found that false memory for intact shapes ("intact"/scrambled > "scrambled"/scrambled) activated LOC at retrieval. Such LOC activity likely reflects conscious shape processing during false memory construction. In the present analysis, we employed the identical fMRI dataset to assess whether scrambled items that were subsequently remembered as intact shapes ("intact"/scrambled) were associated with illusory shape processing at encoding. The conjunction of intact shape processing during encoding (intact shapes > scrambled items) and subsequent false memory for intact shapes (subsequent "intact"/scrambled > subsequent "scrambled"/scrambled) produced activity in LOC. By comparison, the conjunction of shape processing during encoding (intact shapes > scrambled items) and subsequent true memory for scrambled items (subsequent "scrambled"/scrambled > subsequent "intact"/scrambled) did not produce activity in LOC. These findings suggest that false memory for intact shapes was induced by illusory shape processing during encoding. Furthermore, we ran a conjunction of subsequent false memory for intact shapes (at encoding) and false memory for intact shapes (at retrieval) to assess whether retrieval-related false memory activity reflected sensory reinstatement of encoding-related activity. This conjunction revealed overlapping activity in LOC. However, this activity only comprised a subset of LOC activity that was observed at retrieval, which indicates that not all retrieval-related activity reflects sensory reinstatement. The present evidence suggests that retrieval-related activity in LOC during false

memory for intact shapes reflects both sensory reinstatement of illusory shape activity associated with encoding and additional conscious shape processing activity associated with retrieval.

Disclosures: J.M. Karanian: None. **S.D. Slotnick:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.17/VV31

Topic: H.02. Human Cognition and Behavior

Title: Target-lure similarity predicts eye movements during encoding and retrieval in a mnemonic discrimination task

Authors: *D. K. BJORNN[1], C. STRAW[2], E. S. BRIGHTON[2], C. B. KIRWAN[3]
[1]Psychology, [2]Neurosci. Ctr., [3]Psychology and Neurosci. Ctr., Brigham Young Univ., Provo, UT

Abstract: Mnemonic discrimination allows an individual to differentiate very similar memory representations. At encoding, mnemonic discrimination is thought to rely on pattern separation whereby similar memory representations are orthogonalized. At retrieval, false alarms (mistakenly labeling a similar lure as a repeat) are proposed to be due to pattern completion whereby a previously encoded memory representation is reactivated given a noisy or degraded cue. An alternate explanation for false alarms to similar lures is poor encoding of the target stimulus. A recent study by Molitor, Ko, Hussey, and Ally (2014, Hippocampus) used eye tracking as a measure of memory encoding to understand whether discrimination errors come about because of poor encoding or pattern completion. The authors suggested that false alarms to similar lure trials come about because of poor encoding rather than the involvement of pattern completion upon retrieval.
We sought to extend the previous work by using a larger sample size and greater number of trials as well as controlling for lure-pair similarity. Images of everyday objects were displayed one at a time to human participants while eye tracking data were collected. Sixty participants indicated whether each image was an exact repeat, a similar lure, or a novel stimulus. Similarity level for lure trials was controlled so that there was an equal number of high and low similarity lure trials in each block.
Normalized fixation counts were entered into a repeated measures ANOVA with factors for presentation (first, second) and response type (hit, lure correct rejection, lure false alarm). Results supported the finding of Molitor et al. (2014) that lure false alarms had fewer fixations on encoding trials than lure correct rejections, but also diverged from their work by showing the same pattern on retrieval trials. A further analysis was done on lure trials with factors for presentation (first, second), response type (lure correct rejection, lure false alarm), and similarity level (high, low). Low-similarity lure trials showed significantly fewer fixations on encoding

trials for lure false alarms as compared to lure correct rejections. High-similarity lure trials, however, showed significantly fewer fixations on retrieval trials for lure false alarms as compared to lure correct rejections.
These results suggest that inattention at encoding accounts for false alarms to low-similarity lures. However, for high-similarity lures, our data suggest the involvement of pattern completion during retrieval. These results indicate that target-lure similarity is a better predictor of the fixations and response outcome of trials than just poor encoding.

Disclosures: D.K. Bjornn: None. **C. Straw:** None. **E.S. Brighton:** None. **C.B. Kirwan:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.18/VV32

Topic: H.02. Human Cognition and Behavior

Support: Brigham Young University Office of Research and Creative Activities Grant

Title: Mnemonic discrimination in context-dependent memory specificity utilizing eye tracking confidence measures

Authors: *T. WINN[1], A. HEDGES-MUNCY[1], B. KIRWAN[2]
[2]Psychology, [1]Brigham Young Univ., Orem, UT

Abstract: The relationship of objects and their contexts is at the core of long-term declarative memory and critically involves hippocampal processing. To further explore the effect of surrounding visual context on memory for objects, we evaluated behavioral performance in a contextual memory task that especially taxes pattern separation processes. Adding a contextual component to memory specificity tasks is believed to increase the level of difficulty of the task by interfering with the ability to successfully perform pattern separation processes. Some researchers propose that when the object matches its context, there will be more efficient discrimination between stimuli and thus more accurate responses. However, some hypothesize that repeat and similar objects presented with the same background will be more accurately remembered than objects presented with different backgrounds due to the increase of interference. We tested these hypotheses by conducting a mnemonic similarity task in which we manipulated object-background congruency during the encoding phase. We recruited 80 participants to perform the two-part memory task. In the study phase, participants were shown images of everyday objects with backgrounds that were congruent (e.g., a shopping cart against a grocery store) or incongruent (e.g., a gorilla against a classroom). In the test phase, participants were shown images containing items either exactly the same as or similar to the previously studied items (repeats and lures, respectively) but with no background. All images were

presented for 2500 ms with an inter-trial interval of 500 ms. During the test phase, participants were instructed to indicate whether the item in the image was a repeat or lure. As a measure of subject confidence and attention, eye movements were recorded during both phases of the experiment in order to determine time spent studying the object vs. time spent studying the background in the study phase, and time spent studying the object during the test phase.Repeated measures ANOVA showed a difference in performance related to whether the background was congruent or incongruent as related to recognition and discrimination. There was also a relationship between sex and performance of pattern separation tasks. In order to identify possible brain subregions associated with these differences, we intend to continue investigation utilizing functional magnetic resonance imaging (fMRI) techniques.

Disclosures: **T. Winn:** None. **A. Hedges-Muncy:** None. **B. Kirwan:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.19/VV33

Topic: H.02. Human Cognition and Behavior

Title: Exploring the resting state neural activity of monolinguals, late and early bilinguals

Authors: *A. L. HOWELL[1], C. E. GOLD[2], C. B. KIRWAN[1,3]
[1]Neurosci., [2]Spanish Pedagogy, [3]Psychology, Brigham Young Univ., Provo, UT

Abstract: Learning a foreign language is one of many lifestyle factors correlated with a delay in the onset of Alzheimer's disease, a costly public health concern. However, much of the research surrounding bilingualism and memory disorders focuses on individuals who acquire their second language before puberty (early bilinguals); it is unknown whether post-pubertal language acquisition (late bilingualism) affords similar preventative possibilities. This study aimed to ascertain potential differences in memory performance and resting state neural activity for early and late bilingual subjects as compared to monolingual controls.
We recruited ninety young adult subjects (ages 18-30) to one of three groups—early bilingual, late bilingual, and English control—for a total of thirty subjects per group. Bilingual subjects grew up speaking English and Spanish; late bilinguals were native English or Spanish speakers who began learning either Spanish or English, respectively, after puberty. We assessed English and Spanish proficiency with an elicited imitation language task, the results of which are highly correlated with ACTFL's proficiency scale. Additionally, we obtained structural MRI and resting state fMRI data for each participant. No tasks were completed in the scanner.
In addition to scanning, participants completed a mnemonic-discrimination task, which has been shown to be sensitive to hippocampal integrity and related to volume of hippocampal subfields. During the study phase, participants viewed a series of images depicting everyday objects, and

subjectively determined whether each item belonged indoor or outdoor; this judgment facilitated encoding. In the subsequent test phase, participants viewed a second series of images containing images from the original list (repeats) as well as images highly similar to the original images (lures) and entirely new images (foils). For each image, we instructed the participant to make an old/similar/new judgment by pressing specified buttons on the keyboard while the image remained on the screen. The images appeared for a total of 3 seconds and progressed automatically.
We observed significantly greater grey matter volume for late bilinguals compared to controls and early bilinguals in several brain regions including the right hippocampus, left parahippocampal gyrus, and right middle temporal gyrus. Behaviorally, late bilinguals performed significantly better than controls for recognition memory, but not for mnemonic discrimination.

Disclosures: **A.L. Howell:** None. **C.E. Gold:** None. **C.B. Kirwan:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.20/VV34

Topic: H.02. Human Cognition and Behavior

Support: NSF 1422831

Title: Mri data pre-processing steps differentially affect volumetric measures

Authors: *A. HEDGES, N. M. MUNCY, B. KIRWAN
Psychology, Brigham Young Univ., Provo, UT

Abstract: In MRI studies, researchers often clean the data via pre-processing algorithms prior to analyzing registration-based volumes. The various steps in pre-processing algorithms may differentially affect the volumetric output as each pre-processing step adjusts voxel intensities, the mathematical space of the scan, and the overall amount of data available in the scan; such manipulation of the scan may affect the volumetric output. To determine if the pre-processing has a statistical effect on the volume measures, we scanned healthy, young adult participants (n=50) on a Siemen 3T Tim-Trio scanner, with a subset of these participants (n=15) being scanned four subsequent times at the same time each day. All scans were pre-processed following a stepwise pipeline, starting with the scan in original space after a conversion to Neuroimaging Informatics Technology Initiative (NIfTI) format, rotating and cropping the scan, aligning the anterior commissure-posterior commissure (AC-PC) axes, correcting for field inhomogeneity, and finally skull-stripping the scan. After each successive step, we assessed the volumes for the left and right putamen, hippocampus, and middle temporal gyrus (MTG). To test for the effects of pre-processing on volumetric output, we performed a multivariate, repeated

measures analysis on structure volumes following each pre-processing step, correcting for total brain volume. The analysis showed a main effect of pre-processing step and an interaction of step by region-of-interest. Each pre-processing step produced significantly different volumes in the left and right putamen. For the hippocampi, all steps produced significantly different volumes except for the AC-PC alignment and the field inhomogeneity correction. Finally, all steps produced different left and right MTG volumes except for the final skull-stripping step. This indicates that different anatomical regions are differentially affected by the pre-processing steps. We then provide a correction for the pre-processing noise that can be applied in future studies to account for variability introduced by different pre-processing steps.

Disclosures: **A. Hedges:** None. **N.M. Muncy:** None. **B. Kirwan:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.21/VV35

Topic: H.02. Human Cognition and Behavior

Title: The fate of memory representations in mnemonic generalization: An FMRI study

Authors: *N. MUNCY[1], B. KIRWAN[2]
[1]Neurosci., [2]Psychology, Brigham Young Univ., Provo, UT

Abstract: Mnemonic discrimination is the ability to form and retrieve distinct memory representations for similar stimuli or episodes. Recent investigations of mnemonic discrimination have focused on the ability to discriminate between similar items and often induce research participants to generalize and falsely endorse similar lures as repeated (i.e., false alarms). The fate of the underlying memory traces following mnemonic generalization is unknown. Further, the underlying neural processes involved in perpetuating or recovering from a mnemonic generalization are unknown. We therefore developed a protocol to investigate the fate of generalized memory representations, and hypothesized that activation in regions of the medial temporal lobe and prefrontal cortex would predict whether the participant recovered from or perpetuated the mnemonic generalization.Thirty-five healthy, young adults participated in a study-test1-test2 design. During the study phase, participants were exposed to visual stimuli while performing an orienting task. In the test1 phase, participants were exposed to either the same (target) or a similar (lure) stimulus as during the study phase and indicated whether the stimulus was the same or different. The decision that a lure was the same as the original study stimulus is termed mnemonic generalization (and the recognition that it is similar but different is mnemonic discrimination). In the test2 phase, both the target and its lure counterpart were presented, and the participant chose which of the two was the original study stimulus. Study, test1, and test2 were performed inside an MRI scanner while high-resolution (1.8mm3), whole-

brain functional data were collected. We found that neural regions which were differentially activated during encoding predicted subsequent discrimination performance, and that neural regions were differentially activated during test1 discrimination versus generalizations. Critically, and in accordance with our hypothesis, we found a number of regions including the bilateral superior frontal, fusiform, and right middle temporal gyrus that were differentially activated during test1 generalizations that predicted subsequent test2 performance, i.e. regions that predicted whether or not the mnemonic generalization would be perpetuated. We conclude that generalizations can result from poor encoding or poor recovery, and that not all generalizations are equal; that failures to discriminate can result in either multiple memory representations or a rewriting of the original trace, and that the recovery from mnemonic generalization is dependent on the processing of certain anatomic regions.

Disclosures: **N. Muncy:** None. **B. Kirwan:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.22/VV36

Topic: H.02. Human Cognition and Behavior

Title: Physical activity is associated with differential BOLDresponses In the hippocampus during pattern separation: an fMRI study

Authors: *C. B. HODGES, N. M. MUNCY, C. B. KIRWAN
Psychology, Brigham Young Univ., Provo, UT

Abstract: There is growing interest in the impact of physical activity on neural and psychological outcomes, especially given the link between physical activity and improved cognitive functioning in adolescents, young adults, and middle-aged adults. Pattern separation is a computational process associated with declarative memory, and involves the formation of distinct neural representations of incoming stimuli. Pattern separation is often associated with orthogonalizing similar or partially overlapping patterns to reduce memory interference. This present study aims to utilize a mnemonic discrimination paradigm that taxes pattern separation processes to investigate differences in neuronal activity among young adults with various levels of physical activity. It is hypothesized that people with higher levels of physical activity will show pattern separation responses in hippocampal subregions. Sixty right-handed subjects (30 men and 30 women), ranging from ages 18-40, were recruited for one of three groups: a sedentary group, a low-physical activity group and a high-physical activity group. Subjects were given exercise logs and an accelerometer to wear for one week. After this week, subjects had structural (T1- and T2-weighted) and functional (whole-brain high-resolution multi-band) MRI scans. Regions of interest selected were CA1, CA3/dentate gyrus, and subiculum subregions of

the hippocampus. During these MRI scans, subjects completed a continuous recognition mnemonic discrimination task that involved viewing photographs of similar objects, and responding whether or not the image had been shown previously. As hypothesized, subjects with lower levels of physical activity showed decreased BOLD responses, in comparison to the high physical activity group, in the bilateral hippocampal regions during correct responses to our pattern separation task. Additionally, these subjects showed decreased BOLD responses, compared to the high physical activity group, in the medial prefrontal and medial temporal regions for correct responses. These effects were also consistent between the low physical activity and high physical activity group.

Disclosures: C.B. Hodges: None. **N.M. Muncy:** None. **C.B. Kirwan:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.23/VV37

Topic: H.02. Human Cognition and Behavior

Support: 973 Program 2014CB846102,

Title: The neural mechanism of testing effect in episodic memory

Authors: *Z. YE, G. XUE
State Key Lab. of Cognitive Neurosci. and Learning, Beijing, China

Abstract: Retrieval practice, or testing has long been known for its role in benefiting long-term memory performance. Although previous studies have shown that retrieval practice could facilitate the acquisition of new memories and/or the inhibition of competing old memories, the neural mechanisms underlying these effects remain poorly understood. In current study, we applied fMRI and multiple voxel pattern analysis to directly examine the testing effect in memory updating. On day one, subjects were over-trained in words (A)- picture (B) association. On day two, they were introduced the new A-C associations (B and C were of different visual categories) before entering the scanner. In the scanner, the new A-C associations were studied three times under one of the two conditions: restudy vs. testing. Subjects were scanned again on Day Three to test A-C memory performance using a cued recall test. We found that behaviorally, testing could improve A-C recall and reduce the intrusion of A-B memory. MVPA classifier evidence also showed stronger reactivation of target (i.e., C) memory in the medial prefrontal cortex (MPFC) and weaker reactivation of competitor (i.e., B) memories' in the ventral temporal regions. During encoding, the reactivation of target memory increased across repetitions in test condition but decreased in the restudy condition. The left dorsal prefrontal cortex showed greater activation in test condition compared to restudy condition during encoding, which was in turn

associated to better memory updating (target - competitor reactivation) in the MPFC. There were also stronger competitor memory reaction and overall reduced hippocampal activation under the test condition than under the restudy condition. Taken together, our results suggest that testing may facilitate memory updating via a memory reconsolidation mechanisms and the prefrontal cortex plays a critical role in this process.

Disclosures: Z. Ye: None. **G. Xue:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.24/DP13/VV38 (Dynamic Poster)

Topic: H.02. Human Cognition and Behavior

Support: NIH R01-MH062500

Title: Using virtual reality environments to assess context boundary effects and temporal memory performance via spatiotemporal exploration

Authors: *K. M. HORECKA[1,2], M. R. DULAS[1,2], N. J. COHEN[1,2]
[1]Univ. of Illinois at Urbana-Champaign Col. of Liberal Arts and Sci., Urbana, IL; [2]Beckman Inst., Urbana, IL

Abstract: Virtual Reality (VR) has been identified as a useful tool in neuropsychological evaluation thanks to its increased control and measurement capabilities over other methods (Schultheis et al, 2002). More recently, evidence shows that along many measures, VR provides similar ecological validity to real environments (Kuliga et al, 2014). One area of research which is particularly amenable to VR paradigms is learning and memory. Previous work has suggested that context-boundaries impact the binding of sequential information, resulting in within-context information being remembered as closer together than across-context information, even when equidistant, i.e. the segmentation effect (DuBrow & Davachi, 2013). Previous work by our group has shown that segmentation effects can be found in the spatial reconstruction of item locations within and across contexts, with within-context items being placed closer together and across-context items being placed further apart. The present study explored segmentation effects in temporal memory; a VR spatiotemporal navigation task with four contexts (colored time periods) and 2 objects per context (one that appears and one that disappears) was constructed to evaluate how memory for item pair temporal locations within a context differ from pairs which cross contexts. During study, participants were instructed to explore the VR environment and learn the spatiotemporal locations of all items. Importantly, participants were given the ability to explore not only space, but also time, via a button press that changed the direction of the flow of time. Measurements of participant position, orientation, and subsequent memory for item

spatiotemporal locations were assessed across four repeated trials. Results showed significant increases in performance (speed, efficiency of movement/orientation, and memory accuracy) across successive training/test trials. Across all trials, participants showed a significant event segmentation effect, with across-context pairs being remembered as further apart in time and within-context pairs closer together, despite these pairs being equidistant. These segmentation effects remained stable despite improvements in overall accuracy suggesting they are tied to the organization of memory, and not simply a type of error. Lastly, temporal exploration during the first study phase correlated with the size of the segmentation effect, suggesting early exploration behavior may influence the organization of memory relative to context. These results show that temporal segmentation effects are present even when participants can freely navigate the axis of time.

Disclosures: K.M. Horecka: None. **M.R. Dulas:** None. **N.J. Cohen:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.25/VV39

Topic: H.02. Human Cognition and Behavior

Support: NIMH Intramural Research Program: ZIA MH002588-26

Title: Separating memory and attention within the parietal memory network: An fMRI investigation comparing explicit and implicit retrieval processes using overt picture naming and recognition

Authors: *A. W. GILMORE[1], S. E. KALINOWSKI[1], S. C. MILLEVILLE[2], S. J. GOTTS[1], A. MARTIN[3]
[1]Lab. of Brain and Cognition, NIMH/NIH, Bethesda, MD; [2]NIMH / LBC, Bethesda, MD; [3]Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: Differentiating between that which is novel and that which is familiar is a critical aspect of human behavior. This is often accomplished automatically, yet at other times we intentionally reflect upon a specific person or object and attempt to ascertain if we have had any prior experience with them. Prior investigations of such “implicit” and “explicit” retrieval conditions have found their respective neural correlates to be generally distinct.
However, it has been hypothesized that a parietal memory network (PMN) displays broad sensitivity to stimulus novelty or familiarity across numerous task states. PMN regions typically deactivate when orienting to novel, but activate when orienting to familiar, stimuli. To further understand the functional significance of this network, we must be able to separate activity that is related to familiarity *per se* from activity that may be driven by attentional demands which differ

across task states.
We therefore designed a task in which subjects would encounter novel and familiar stimuli in both implicit and explicit retrieval conditions, matching the two as closely as possible. While undergoing fMRI, subjects were presented with images of animals and man-made objects and either named the object aloud or verbally made an old/new recognition judgement. Naming was performed in the context of only novel items (as in typical “encoding” conditions), or was done in a mix of old and new items (as in typical “retrieval” conditions). Recognition was always performed on a mix of old and new items. A slow event-related design helped ensure that speaking-related motion did not corrupt peak response estimates, and a multi-echo BOLD sequence was employed to help identify and remove noise components within each scan run. Task-related differences between novel and repeat items were identified broadly across the cortex, reflecting both increases and decreases across repetitions. Notably, regions within the PMN demonstrated robust old > new effects in both the naming and recognition tasks. This is consistent with the hypothesis that activity within PMN regions is sensitive to familiarity in general. However, we also observed that activity was greater during recognition than during naming for familiar items, suggesting a prominent role for attentional or task demands in determining PMN responses. Collectively, these findings suggest that the basic old > new effects which are so consistently observed in the network cannot be fully understood without consideration of the task states in which they appear.

Disclosures: **A.W. Gilmore:** None. **S.E. Kalinowski:** None. **S.C. Milleville:** None. **S.J. Gotts:** None. **A. Martin:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.26/VV40

Topic: H.02. Human Cognition and Behavior

Support: NSFC Project (81371631)

NSFC Project (81422024)

Beijing Nova Program (Z141110001814068)

Thousand Youth Talents Plan (Y4HX072006)

Hundred Talent Program of Chinese Academy of Sciences (Y3CX022003)

Title: The stimulus specific representation underlies similar memory discrimination

Authors: *Z. GAO[1,2], L. WANG[1]
[1]Inst. of Psychology, Chinese Acad. of Sci., Beijing, China; [2]Univ. of Chinese Acad. of Sci., Beijing, China

Abstract: When some features of our experiences overlap, successful discrimination among similar memories involves in remembering the specific details that can dissociatively characterize prior experience. This ability builds up formation of event-unique memory traces termed as pattern separation that mainly rely on the hippocampus, but are proposed to occur throughout the brain. However, we have little knowledge on neural representation in similar-item memory discrimination. In this study, participants viewed images of objects (each with 3 repetition) while performing a likeness judgement task during fMRI scan; then, in a later recognition test, they judged whether test items (3 categories: target: same as studied item, foil: novel, completely different with studied item; lure: similar but not same with studied items) were 'same' or 'new' relative to the items seen before. Using representational pattern analysis, we identified a set of regions in which neural representation fidelity during encoding can predict memory discrimination correct (lure judged as new) or false (lure judged as same). Especially, we only found that representation fidelity in the MTL are higher for correct than false lure memory. Besides, the MTL showed higher Item specificity (within item similarity minus between item similarity) for correct than false lure memory discrimination. Furthermore, encoding-retrieval similarity analysis revealed that successful memory discrimination was accompanied by greater item level reinstatement in MTL, ITG, precuneus, and frontal orbital cortex.

Disclosures: Z. Gao: None. **L. Wang:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.27/VV41

Topic: H.02. Human Cognition and Behavior

Support: James S. McDonnell Scholar Award

Title: The influence of prediction error on episodic memory reconsolidation

Authors: *A. H. SINCLAIR, M. D. BARENSE
Psychology, Univ. of Toronto, Toronto, ON, Canada

Abstract: A reminder can temporarily destabilize an established memory trace, creating a window of opportunity during which the memory can be modified, until it is stabilized again through the process of *reconsolidation*. Recent research has suggested that a *prediction error* at

the time of reactivation is necessary to destabilize a memory trace. The present study was the first to use video stimuli to investigate the influence of prediction error on the reconsolidation of complex episodic memories. On Day 1, participants (N=23) viewed 18 target videos, each depicting a salient action-outcome event (e.g., a baseball batter hitting the ball out of the park). On Day 2, we reactivated memories by presenting each target video again either in *match* form, identical to the original viewing, or *mismatch* form, cut off abruptly before the completion of the video (e.g., the baseball batter cut off mid-swing). We hypothesized that mismatch-reactivation would generate a prediction error, most effectively destabilizing memories. Participants also viewed 18 new *interference* videos, each semantically-related to one of the target videos (e.g., a baseball fan in the stands catching a foul ball). Critically, participants in the Experimental condition received the reactivation prior to interference, whereas participants in the Control condition received the interference prior to reactivation. On Day 3, participants completed an interview-style recall test on the target videos. The rate of *intrusions*, details from the interference videos mistakenly attributed to the corresponding target videos, was significantly higher in the Experimental condition, $F(1, 21) = 20.11$, $p < .001$, $\eta^2 = 0.5$. Furthermore, there was a significant interaction between condition and reactivation type, $F(1, 21) = 5.23$, $p = .033$, $\eta^2 = .2$. In the Experimental condition, mismatch reactivation produced a higher rate of intrusions than match reactivation. In contrast, reactivation type did not affect intrusions in the Control condition. Self-reported confidence was rated to be “moderately high” for all participants and did not differ between conditions, suggesting that metamemory judgments do not accurately reflect memory distortions.

Our results provide a compelling demonstration that episodic memories undergo reconsolidation. In particular, the intrusions from the interference videos characterize the reconsolidation process as a dynamic updating mechanism which allows new information to be incorporated into a memory. Moreover, we point to a recently proposed mechanism, implicating prediction error in the destabilization and reconsolidation process.

Disclosures: **A.H. Sinclair:** None. **M.D. Barense:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.28/VV42

Topic: H.02. Human Cognition and Behavior

Support: NWO grant 446-14-009

NIH grant MH048832

Harvard Catalyst grant TR001102

Title: Sleep selectively enhances associative aspects of emotional memories

Authors: *R. COX[1,3], M. VAN BRONKHORST[2], H. GOMILLION[2], A. C. SCHAPIRO[4], R. STICKGOLD[5]
[1]Dept. of Psychiatry, [2]Beth Israel Deaconess Med. Ctr., Boston, MA; [3]Psychiatry, Harvard Med. Sch., Boston, MA; [4]Psychiatry, Beth Israel Deaconess Med. Ctr. / Harvard Med., Boston, MA; [5]Dept Psychiatry, Ctr. For Sleep and Cognition, Boston, MA

Abstract: Introduction Sleep promotes the retention of episodic memories, but prioritizes memory stabilization depending on items' perceived relevance. Typically, emotional material benefits more from a period of sleep than neutral information. At a more fine-grained level, however, it is unclear which aspects of emotional memory are preferentially consolidated. Here, we test whether selective consolidation effects differ for item recognition and associative memory.
Methods Forty-five healthy volunteers encoded 128 neutral and 128 negative stimuli presented visually to either the left or right visual field, with separate groups encoding in the morning (n=16) and evening (n=29). Immediately after encoding, and after a 12 h interval containing either sleep or wake, individuals were shown items that were presented before as well as new items. Crucially, items were now presented centrally, not lateralized. Subjects judged each item in two distinct ways. First, to measure item recognition, subjects provided an old/new rating. Second, to measure associative memory aspects, they indicated the side where they believed the item was originally presented. We analyzed behavioral data by performing several 2 (GROUP: sleep/wake) x 2 (VALENCE: emotional/neutral) x 2 (TIME: immediate/delayed) mixed ANOVAs. In addition, we recorded high-density EEG in the sleep group throughout the encoding, retrieval, and sleep periods, to search for neural correlates of sleep-dependent memory effects.
Results Source memory for correctly recognized old items was selectively enhanced for emotional items over a period of sleep vs. wake. This was expressed as a significant 3-way interaction between all factors (P=0.004). Follow-up tests demonstrated that while associative memory performance decreased across time for emotional (P=0.015) and neutral items (P=0.10) in the wake group, and for neutral items in the sleep group (P=0.001), source memory improved significantly for emotional items across sleep (P=0.035). In contrast, changes in recognition memory over time did not depend on whether subjects slept or not, either for emotional or neutral items. EEG oscillatory correlates underlying behavioral consolidation effects will be presented.
Conclusion These results of selective strengthening of associative links for emotional items during sleep are consistent with theoretical accounts of sleep's role, recoding labile, hippocampus-dependent, relational memories to a more stable neocortical format.

Disclosures: R. Cox: None. **M. van Bronkhorst:** None. **H. Gomillion:** None. **A.C. Schapiro:** None. **R. Stickgold:** None.

Poster

431. Memory Encoding and Retrieval Processes

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 431.29/VV43

Topic: H.02. Human Cognition and Behavior

Support: NIH Grant MH048832

Title: Strength of initial encoding in the selection of memories for sleep-dependent consolidation

Authors: *D. DENIS[1], R. STICKGOLD[2]
[1]Ctr. for Sleep and Cognition, Boston, MA; [2]Dept Psychiatry, Ctr. For Sleep and Cognition, Boston, MA

Abstract: A large body of research has shown that sleep plays an important role in the consolidation of recently acquired memories. However, the brain does not retain all of the information that it encodes each day. Recent research has started to uncover the mechanisms by which the brain selects which information should be preserved and therefore subsequently consolidated during sleep, and which information is to be forgotten. The aim of this study was to investigate the role that initial encoding strength has on subsequent sleep-dependent consolidation. Previous studies have found mixed results, with some evidence showing that weaker memories are prioritized for consolidation during sleep, with others showing that stronger memories are prioritized. Here we had participants learn unrelated concrete noun word-pairs to 5 different levels of encoding strength. After learning, there was a 5-minute eyes-closed quiet rest period followed by a recall test of all word pairs (test 1). Participants were then divided into 3 groups and were told to return for a re-test (test 2) at either 4 hours (short-wake), 12 hours (long-wake), or 24 hours (sleep) later. Change scores in recall performance was calculated as the difference in the number of pairs recalled at test 1 and test 2. Preliminary results suggest a beneficial effect of sleep on weaker encoded memories, but the absolute weakest level of encoding showed no sleep-dependent benefit. Strongly encoded memories did not benefit from a period of sleep. These results show that the brain prioritizes weaker memories for consolidation during sleep, but a minimum threshold has to be reached in terms of initial encoding strength in determining which memories to select for consolidation. Very weak memories are not selected possibly because the memory trace is too poor for consolidation to be possible. On the other hand, strongly encoded memories are also not selected, possibly because the memory trace is strong enough that further consolidation of the memory is not necessary for long-term retention.

Disclosures: D. Denis: None. **R. Stickgold:** None.

Poster

432. Visual Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 432.01/VV44

Topic: H.02. Human Cognition and Behavior

Support: Wellcome Trust 106926

Title: Neural consequences of directing retrospective attention in visual working memory

Authors: *P. M. BAYS, R. TAYLOR
Dept. of Psychology, Univ. of Cambridge, Cambridge, United Kingdom

Abstract: An informative cue that directs attention to one of several items in working memory improves subsequent recall of that item. Here we examine the mechanism of this retro-cue effect using a model of short-term memory based on neural population coding. Our model describes recalled feature values as the output of an optimal decoding of spikes generated by a tuned population of neurons. This neural model is found to provide a better account of human recall data than an influential model (Zhang & Luck, Nature, 2008) that assumes errors can be described as a mixture of normally-distributed noise and random guesses. The retro-cue benefit is revealed to be a consequence of a relative increase in the firing rate of the encoding population, with no change in tuning specificity (Figure 1). Additionally, a retro-cued item is less likely to be swapped with another item in memory, an effect that can also be explained by increased activity of the underlying population. These results provide a parsimonious account of the effects of retrospective attention on recall and demonstrate a powerful method for probing neural representations with behavioural tasks.

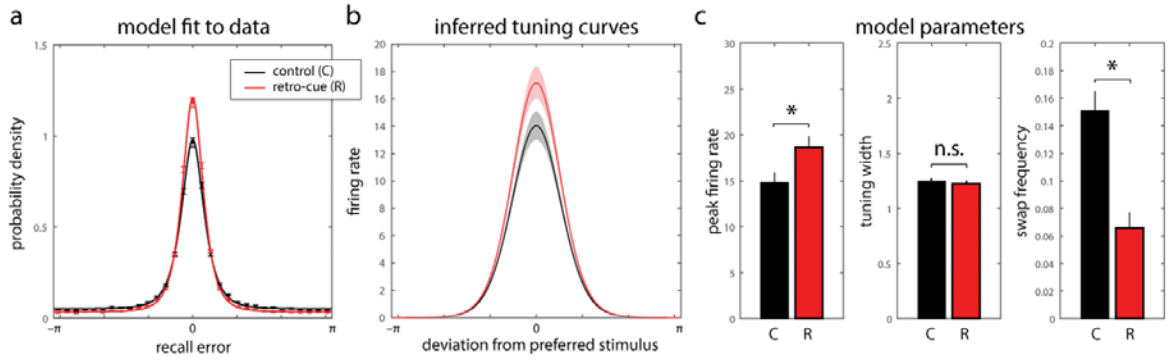


Figure 1. (**a**) Symbols indicate empirical recall errors (mean ± SE) and curves show fit of population coding model, for trials with (red) and without (black) a retro-cue. (**b**) Inferred tuning functions of neurons encoding the visual memoranda. (**c**) Model parameters: peak firing rate and swap frequency were modulated by retro-cueing, with no effect on tuning width.

Disclosures: P.M. Bays: None. **R. Taylor:** None.

Poster

432. Visual Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 432.02/VV45

Topic: H.02. Human Cognition and Behavior

Support: NIH Grant MH064498

Title: Maintenance of location information for the unattended memory item during visual working memory

Authors: *Q. YU, B. POSTLE
Univ. of Wisconsin-Madison, Madison, WI

Abstract: Previous studies of information held at different levels of priority in visual working memory have suggested that only information in the current focus of attention is represented in an active state. The “unattended memory item (UMI)”, on the other hand, may be retained in an “activity-silent” state. In the current study, we aimed to investigate how the system keeps a record of a UMI while it is still potentially relevant for behavior. Participants viewed two gratings with different orientations (one in each hemifield) and were asked to remember both of them across an initial delay (Delay 1). In the Retain-One-Item (R1) condition, participants saw a cue indicating which of the two items would be relevant for the remainder of the trial, and were tested on their memory for that item after a second delay (Delay 2), then re-cued and re-tested on that same item after a third delay (Delay 3). In the Retain-Two-Items (R2) condition, participants performed a similar task, except that the first cue did not predict which item would be relevant for the final memory probe. Because the cued and uncued items were separated in space, we could test whether information about the retinotopic location of the UMI might be retained across Delay 2, using multivariate pattern analysis (MVPA). Independently, we could track the representation of orientation with inverted encoding models (IEM). In the R2 condition, consistent with previous work, we observed degraded neural representation of the orientation of the UMI after the first cue. MVPA, however, indicated that the location of the UMI, but not the attended memory item, was robustly retained across Delay 2. The comparable location information could not be decoded for either item in the R1 condition. These patterns held across multiple regions in early visual cortex. These results support the idea that the brain may retain an active representation of episodic information about UMIs so long as they remain potentially relevant for behavior.

Disclosures: Q. Yu: None. **B. Postle:** None.

Poster

432. Visual Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 432.03/VV46

Topic: H.02. Human Cognition and Behavior

Support: MH064498

Title: The neural representation of stimulus information, of the stimulus location, and of the location-dependence of stimulus information in visual working memory

Authors: *Y. CAI[1], D. A. SHELDON[2], B. R. POSTLE[2]
[1]Beijing Normal Univ., Beijing, China; [2]Univ. Of Wisconsin–Madison, Madison, WI

Abstract: This study applied multivariate inverted encoding modeling (IEM) to fMRI data to track the dynamics of stimulus representation across a visual working memory (VWM) task. Each trial began with the presentation of an oriented bar ("sample") in one of four locations (one in each quadrant), followed by a blank 8-sec delay, followed by a recall dial requiring estimation of the sample orientation. The recall dial appeared in the same location as the sample. Across 11 subjects, mean estimation error our 6.7 deg., and a 2-factor mixture model estimated a mean guess rate of less than 5%. Sample-evoked BOLD signal intensity was elevated in occipital, parietal, and frontal regions; delay-period activity in parietal and frontal. Unilateral ROIs were defined in each of these lobes as the 400 voxels showing the greatest difference in sample-evoked activity for contralateral versus ipsilateral stimuli. When IEMs were trained and tested at each TR, in all three regions, stimulus orientation could be reconstructed with signal evoked by contralaterally presented samples as well as by signal evoked by ipsilaterally presented samples. In the occipital ROIs, the same was true for data from the delay and recall periods. In the parietal ROIs, orientation could be reconstructed during the delay period only when the sample had been presented contralaterally. These stimulus representations were dynamic, because reconstruction failed when sample-trained IEMs were fed data from later portions of the trial. To further investigate the temporal evolution of these memory representations, we also trained IEMs on signal from the occipital ROIs from the delay and the recall periods. Whereas recall-period output of sample-trained IEMs distinguished those that had been trained contralaterally from those that had been trained ipsilaterally, information about sample location was weaker in delay-trained IEMs, and weaker still in probe-trained IEMs, suggesting that the mnemonic representation of orientations became progressively less location-dependent over time. In contrast to this, multivariate pattern analysis (MVPA) of the same ROI indicated that occipital cortex retained a robust representation of sample location through the trial. These findings demonstrated that the neural representation of the location dependence of nonspatial stimulus features can be dissociated from the neural representation of stimulus location, per se.

Disclosures: **Y. Cai:** None. **D.A. Sheldon:** None. **B.R. Postle:** None.

Poster

432. Visual Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 432.04/VV47

Topic: H.02. Human Cognition and Behavior

Support: JSPS KAKENHI Grant Number JP16H01672

Title: Neural substrate of objects' material properties held in visual working memory

Authors: *M. FUJIMICHI, H. TSUDA, H. YAMAMOTO, J. SAIKI
Kyoto Univ., Kyoto-Shi, Japan

Abstract: Previous studies on neural mechanisms underlying visual working memory have shown that intraparietal sulcus plays important roles for the memory of color and shapes. In contrast, studies on perception of objects' material properties reported activities in the ventral visual pathway. Which regions are responsible for visual working memory for objects' material properties? In the present study, we conducted an fMRI experiment to explore this question. In the main task, participants performed a delayed discrimination task regarding objects' surface glossiness or roughness in a 3T scanner. In each trial, two sample objects were sequentially presented, followed by a numerical cue about which objects' material property to memorize. When the two samples were different in glossiness (roughness), the participants had to memorize the glossiness (roughness). After an 11s delay, a probe object was presented and the participants indicated which one of the probe object and the memorized one had higher glossiness (lower roughness). The imaging runs of the working memory task were followed by two kinds of imaging: localizer runs to identify brain regions processing objects, color, faces, and scenes, and phase-encoded retinotopy measurements to define retinotopic visual areas. To explore working memory representations for the objects' material properties, we conducted a multi-voxel pattern analysis (MVPA) to predict the memorized material property, glossiness or roughness. The results of MVPA during the delay period showed above-chance accuracies in the ventral object vision pathway and intraparietal sulcus. In contrast, early visual areas could not predict the memorized material property. These results suggest that the ventral object vision pathway and intraparietal sulcus, but not early visual areas, can carry information about objects' material properties even though there are no physical stimulus.

Disclosures: **M. Fujimichi:** None. **H. Tsuda:** None. **H. Yamamoto:** None. **J. Saiki:** None.

Poster

432. Visual Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 432.05/VV48

Topic: H.02. Human Cognition and Behavior

Support: NSFC 81371631, 81422024

Beijing Nova Program Z141110001814068

Thousand Youth Talents Plan Y4HX072006

Hundred Talent Program of CAS Y3CX022003

Title: Neural oscillations underlie maintenance of simultaneously presented multi-items in visuospatial working memroy

Authors: *Y. PAN[1,2], C. QIN[1,2], Z. TAN[1,2], L. REN[3], W. ZHOU[4], L. WANG[1,5,2]
[1]Inst. of Psychology, CAS Key Lab. of Mental Hlth., Beijing, China; [2]Univ. of Chinese Acad. of Sci., Beijing, China; [3]Beijing Key Lab. of Neuromodulation, Beijing Inst. of Functional Neurosurgery, Xuanwu Hospital, Capital Med. Univ., Beijing, China; [4]Epilepsy Ctr. of Yuquan Hospital, Tsinghua Univ., Beijing, China; [5]CAS Ctr. for Excellence in Brain Sci. and Intelligence Technol., Shanghai, China

Abstract: Working memory (WM) is a buffer storage in mind, consisting of verbal and visual forms. Although the former has had been studied well that we could hold several verbal stimuli as chunks in mind, the study of multi-object visual working memory (VWM) attracted some controversies. The present study used intra-cranial electroencephalography (iEEG), recorded neural activity of neuronal population directly from 30 refractory epileptic patients, to explore how our brain maintains multiple-items simultaneously in mind. On studying of multi-objects maintenance, memory load is a very commonly used manipulation. While in traditional paradigms, memory load is indexed by the number of items in display. This kind of experiments certainly involves a confounding factor, namely perceptual load. Here, we developed a novel visuospatial delayed match to sample task (DMST) to separate these two factors. During sample display, all eight spatially dots (non-colorized and colorized dots) presented simultaneously. But only the number of colorized ones (one, two or four dots), rather than the number of total dots (eight), denoting the memory load. Therefore, the perceptual load (the number of all dots) kept the same when manipulating memory load. Preliminary results of this study indicated that behavior performance deteriorated severely with increasing memory load. And the capacity estimator, Cowan's K value first increased then saturated at load 2 condition, which in consistence with the idea that visual working memory was severely limited. Event related

potentials (ERPs) were analyses to validate the effectiveness of perceptual load control. And no load effect was observed during the first 320 ms perceptual period after sample onset, which contained the evoked response components P1 and N1. For the recorded local field potentials (LFPs), time-frequency decomposition with Morlet wavelet was conducted on each channel. Then ANOVA of load effect was analyzed with cluster correction on these time-frequency representations to find the working memory task related channels. The result manifested that these channels distributed widely across different recorded cortical cortexes, namely frontal, parietal, temporal and occipital region. In addition, the number of significant channels in different cortexes exerted no differences under the Mantel-Haenszel test. Therefore, we concluded that when maintain multi-items simultaneously in mind, different cortex regions work together as a network. And how does these regions communication will be investigated in the later analyses.

Disclosures: **Y. Pan:** None. **C. Qin:** None. **Z. Tan:** None. **L. Ren:** None. **W. Zhou:** None. **L. Wang:** None.

Poster

432. Visual Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 432.06/VV49

Topic: H.02. Human Cognition and Behavior

Support: NEI T32-EY007136

NIH R01-EY016407

Title: Resource allocation and confidence in visual working memory

Authors: *A. H. YOO[1], Z. KLYSZEJKO[1], W. J. MA[1,2], C. E. CURTIS[1,2]
[1]Psychology, [2]Ctr. for Neural Sci., New York Univ., New York, NY

Abstract: To improve the precision of working memory (WM), people flexibly allocate attentional resources across items prioritized by their behavioral relevance (Klyszejko et al., 2014; Emrich et al., 2017). Moreover, people are less confident when they have a noisier WM representation (Rademaker et al., 2012). While past computational models of WM typically account for varying item precision through set size manipulations, they do not account for strategies that distribute WM resources to a fixed number of items according to their priority, nor do they account for memory confidence. Here, we collected psychophysical data from two experiments and extended the variable precision (VP) model of WM (van den Berg et al., 2012) to address two aims: how the allocation of WM resources reduces WM error and whether a VP model could account for the trial-to-trial association between WM error and confidence. In Exp.

1, participants performed a memory-guided saccade task in which we manipulated priority by varying the probability that each of four targets would later be the saccade goal. Behaviorally, errors and response times (RT) decreased monotonically with increasing priority. Through computational modeling, we demonstrate that participants did not allocate resources proportional to priority. Rather, they slightly under-allocated to high and over-allocated resources to low priority targets. In Exp. 2, following the memory-guided saccade, participants indicated their memory confidence by adjusting the size of a disc centered on their saccade landing. A smaller disc led to a higher reward, but only if the target was within the disc. Again, errors and RT decreased with priority. Additionally, disc size decreased with priority and was positively correlated to WM error. We extended the computational models from Exp. 1 to include confidence reports, defined an optimal observer model, and compared which model fit each participant's data best. We replicate the results of Exp. 1; the best fitting model suggests that participants under-allocate to high and over-allocate resources to low priority targets relative to proportional or optimal. The best-fitting model also captures the relationship between WM error and disc size. These results demonstrate that, while not optimally or even proportionally, people allocate resources based on priority to enhance performance. Moreover, people are aware of the fidelity of their WM, which they perhaps use to temper memory-guided responses.

Disclosures: **A.H. Yoo:** None. **Z. Klyszejko:** None. **W.J. Ma:** None. **C.E. Curtis:** None.

Poster

432. Visual Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 432.07/VV50

Topic: H.02. Human Cognition and Behavior

Title: Retroactive spatial prioritization in working memory

Authors: K. MAXOOD, A. TEMUDO, *K. K. SREENIVASAN
New York Univ. Abu Dhabi, Abu Dhabi, United Arab Emirates

Abstract: Working memory (WM) is famously capacity limited. A key feature of WM is that its resources can be deployed flexibly in order to prioritize individual items over others, thus increasing the precision with which these items are maintained and recalled. This has most notably been demonstrated using a retrocuing paradigm, where performance is enhanced by a cue during the memory delay that indicates which memory item will be tested. Other recent demonstrations of the flexible allocation of WM resources come via studies that manipulate attentional priority during encoding, either by varying the bottom-up salience or the top-down behavioral relevance of the different items in the memory display. As a result, higher priority items are maintained with higher precision, while lower priority items are maintained with lower precision. An open question is whether attentional prioritization can occur during WM

maintenance, after encoding is complete.
We sought to address this question by having subjects perform a modified delayed estimation task for spatial WM where spatial priority was indicated by a retrocue. On each trial, subjects saw four dots presented on an invisible annulus and retained the location of these dots over a brief memory delay. In the middle of the memory delay, subjects were presented with a central retrocue that indicated the probability (either 0, 0.1, 0.3, or 0.6) that each of the four locations would be probed at the end of the trial. At the end of the memory delay, the subjects were cued to report the location of one of the items. The critical test was whether the degree of error in subjects' reports was proportional to the probed item's rank ordered probability as indicated by the retrocue. We predicted that the amount of response error would be least for items that indicated a 0.6 probability of being probed, and would increase as the probability of being probed decreased.
In line with our prediction, there was a roughly linear effect of priority on average memory error, with the least error for the item cued with 0.6 probability and the most error for the item cued with 0.1 probability. This graded effect provides evidence that WM representations can be assigned a rank-ordered priority after encoding. In addition to providing evidence that mechanisms of spatial prioritization generalize to WM maintenance, these findings have important implications for models that posit multiple representational states in WM.

Disclosures: K. Maxood: None. **A. Temudo:** None. **K.K. Sreenivasan:** None.

Poster

432. Visual Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 432.08/VV51

Topic: H.02. Human Cognition and Behavior

Support: NIH R01-EY016407 (CC, TCS, MR)

NIH T32-EY007136-23 (TCS)

NIH R01-EY020958 (WM, AY)

Nvidia Hardware Grant (TCS)

Title: Decoding uncertainty in visual spatial short term memory from retinotopic cortex

Authors: *T. C. SPRAGUE, M. RAHMATI, A. YOO, W. MA, C. E. CURTIS
Dept. of Psychology, New York Univ., New York, NY

Abstract: When performing tasks requiring the maintenance of information over a brief delay period, observers can introspect the quality of visual short-term memory (VSTM) representations

to optimally guide behavior (Rademaker et al, 2012; Suchow et al, 2016). According to theories of Bayesian probabilistic population coding, such memory representations are manifest as probability distributions over feature values, and the width of these distributions reflects uncertainty in the represented feature value (Ma et al, 2006). Accordingly, if VSTM representations are maintained using probabilistic population codes, and they can be accurately modeled and sensitively measured, representation accuracy as indexed by neural decoding error and behavioral recall precision should be worse when decoded uncertainty of neural representations is greater. We tested these predictions by assaying neural representations of remembered spatial positions and their uncertainty across visual, parietal, and frontal cortex during a memory-guided saccade task using functional MRI. On each trial, participants precisely maintained the position of a briefly-flashed target stimulus over a 10 s delay interval, then generated a memory-guided saccade to the remembered position. We adapted a recently-developed multivariate decoding method based on a Bayesian generative model (van Bergen et al, 2015), which estimates a probability distribution over feature values given neural activation patterns to recover estimates of represented spatial positions and their uncertainty on each trial. In several topographic visual maps, decoding errors increased with increasing decoded uncertainty, consistent with predictions from probabilistic population coding. Additionally, when decoded uncertainty was high during the memory delay, subsequent memory-guided saccades tended to be less accurate. A parsimonious cortical encoding scheme for spatial VSTM representations could exploit the retinotopic organization across much of human cortex, with the topography of feature-selective activation during spatial VSTM maintenance matching the retinotopic structure of visual maps (Mackey, Winawer & Curtis, 2017). Indeed, during the delay period, position preferences of voxels exhibited strong topographic organization along the cortical surface, and this relationship was especially strong in regions which representations related to behavioral performance. Together, these results demonstrate probabilistic population codes implemented across retinotopic maps as a plausible mechanism for the maintenance of visual spatial positions over a delay, as well as their uncertainty.

Disclosures: **T.C. Sprague:** None. **M. Rahmati:** None. **A. Yoo:** None. **W. Ma:** None. **C.E. Curtis:** None.

Poster

432. Visual Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 432.09/VV52

Topic: H.02. Human Cognition and Behavior

Support: NSF BCS 1558535

NSF OIA 1632849

Title: Oculomotor capture reveals trial-by-trial neural correlates of attentional guidance by contents of visual working memory

Authors: *V. BECK, T. VICKERY
Dept. of Psychological and Brain Sci., Univ. of Delaware, Newark, DE

Abstract: Evidence from attentional and oculomotor capture, contingent capture, and other paradigms suggests that mechanisms supporting human visual working memory (VWM) and visual attention are intertwined. Features held in VWM bias guidance toward matching items even when those features are task irrelevant. However, the neural basis of this interaction is underspecified. Prior examinations using fMRI have primarily relied on blocked designs or coarse comparisons across experimental conditions that produce varying amounts of capture. To examine the neural dynamics of attentional capture on a trial-by-trial basis, we applied an oculomotor paradigm that produced discrete measures of capture. Hollingworth, Matsukura, and Luck (2013) previously demonstrated that VWM-matching items attract attention in an oculomotor response task. On each trial, subjects were shown a memory item, followed by a blank retention interval, then a saccade target that appeared to the left or right. On some trials, an irrelevant distractor appeared above or below fixation. Once the saccade target was fixated, subjects completed a forced-choice memory test. Critically, either the target or distractor could match the feature held in VWM. Although task irrelevant, this manipulation produced differences in behavior: participants were faster to saccade to a VWM-matching target, and more likely to saccade first to an irrelevant VWM-matching distractor - providing a discrete measure of capture. We replicated these findings while recording eye movements and scanning participants' brains using fMRI. To examine the neural basis of oculomotor capture, we separately modeled the retention interval for capture and non-capture trials within the distractor-match condition. We found that a region previously associated with VWM, dorsolateral prefrontal cortex, was more active during the retention interval of capture than non-capture trials, suggesting that active maintenance of target information in VWM differentially predicted the tendency to be inappropriately guided to the VWM-matching distractor. In addition, we modeled eye movement latency within each condition. For target-matching trials, with or without a distractor, longer latency positively predicted left hippocampal activity, compared to target non-match trials. These findings suggest that more efficient long-term storage of the target feature mitigated speeding of eye movements, selectively for target-matching conditions. Our findings demonstrate the power of trial-by-trial analyses of oculomotor capture as a means to examine the underlying relationships between VWM and attentional guidance systems.

Disclosures: V. Beck: None. **T. Vickery:** None.

Poster

432. Visual Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 432.10/VV53

Topic: H.02. Human Cognition and Behavior

Support: NSERC

Title: Trans-saccadic integration and visual working memory

Authors: *A. L. FROST[1], M. NIEMEIER[2]
[1]Psychology, Univ. of Toronto, Toronto, ON, Canada; [2]Univ. Toronto, Scarborough, ON, Canada

Abstract: The eyes make about three fast movements per second, and across these “saccades” the brain needs to update the spatial locations of objects to maintain a stable representation of the world. This is achieved through trans-saccadic integration, involving processes of spatial remapping together with memory buffers. To understand how these processes relate to other cognitive mechanisms, in the current study we measured people’s trans-saccadic and other types of working memory performance. In the trans-saccadic task participants briefly viewed arrays of objects, followed by a saccade and then used a mouse cursor to click on one of the remembered object locations. In addition, they performed an n-back and a change detection task, two well-established spatial visual working memory paradigms. We found that both classic measures were unrelated to systematic errors during the trans-saccadic task. However, unsystematic errors were predicted by n-back performance. Surprisingly, working memory capacity as quantified with change detection showed no correlations. Follow-up studies altered the change detection task in several ways to identify the features that would make it more similar to the other tasks. We ruled out features such as interference and remapping locations across visual fields. In contrast, forcing participants to detect changes based on egocentric rather than allocentric information did produce significant correlations. Our results suggest that trans-saccadic integration involves separate mechanisms for trans-saccadic remapping of spatial information and trans-saccadic memory. Moreover, trans-saccadic memory relies on similar buffers as other forms of working memory depending on the spatial coordinate system, with separate buffers for egocentric and allocentric representations of space.

Disclosures: **A.L. Frost:** None. **M. Niemeier:** None.

Poster

432. Visual Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 432.11/VV54

Topic: H.02. Human Cognition and Behavior

Support: NSF GRFP (DGE 1342436)

NIH R01 EY016407

Title: Frontal visual field maps mediate noise resilience of working memory

Authors: *W. MACKEY[1], C. E. CURTIS[2]
[1]New York Univ., New York, NY; [2]Psych & CNS, NYU, New York, NY

Abstract: Working memory (WM) extends the temporal period within which neural representations can be integrated and transformed, enabling a vast array of cognitive abilities. Conversely, WM has severe capacity limitations, and varies widely between individuals and across the lifespan (Anders, Fozard, & Lillyquist, 1972). Psychophysical studies and computational models indicate that random noise corrupts the quality of WM representations (Bays, 2015). Recently, we reported that the size of visual field maps in frontoparietal and early visual cortex predicted individual differences in WM precision (Curtis, et al., SFN 2016). Moreover, we simulated the fidelity of WM in various sizes of neural networks and found that smaller simulated populations were noisier. These findings suggest that WM resource limitations may have a structural basis. However, these relationships were correlational in nature, and could not address whether this relationship was unique to either encoding or maintenance processes. To address these limitations, we combined computational neuroimaging and repetitive transcranial magnetic stimulation (TMS) during a classic spatial WM task to investigate the causal relationship between individual differences in structural properties of visual field maps in frontal cortex and WM precision. First, we used population receptive-field mapping (Mackey, Winawer, & Curtis, 2017) to identify a visual field map that lies in the superior branch of the precentral sulcus (sPCS), a frontal area necessary for WM (Mackey et al., 2016). This served as our stimulation target in each subject. Second, we measured the effect on memory accuracy of a short burst of TMS applied during the delay period of a memory-guided saccade task. We assumed that TMS would introduce random noise, perturbing the population activity in sPCS (Mackey & Curtis, 2017). We predicted that sPCS map size would mediate the effects of TMS on WM performance. Indeed, we found a relationship between individual differences in map size and the TMS induced decrement in WM precision. Specifically, TMS applied during the retention interval caused a greater reduction in WM accuracy in subjects with smaller sPCS maps. Interestingly, subjects with large maps were resilient and were hardly affected by TMS. Together, these results indicate that 1) the sPCS is necessary for maintaining spatial WM, 2)

sPCS size may place a hard constraint on WM resources, and 3) individual differences in sPCS size may predict one's resilience or the degree to which WM representations are corrupted by noise.

Disclosures: W. Mackey: None. **C.E. Curtis:** None.

Poster

432. Visual Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 432.12/VV55

Topic: H.02. Human Cognition and Behavior

Support: Wellcome Trust Grant 106926

Title: Efficient coding in visual working memory accounts for stimulus-specific variations in recall

Authors: *R. TAYLOR, P. M. BAYS
Dept. of Psychology, Univ. of Cambridge, Cambridge, United Kingdom

Abstract: Human recall errors in visual working memory (VWM) show systematic variation within feature dimensions. For example, recall of color appears biased toward certain color prototypes, and recall of orientation is more precise for cardinal than oblique directions. While a number of existing models can approximate the average distribution of recall error across target stimuli, attempts to capture the way in which error varies with the choice of target have been ad hoc. Here we build on an existing account of the neural architecture of VWM, in which stimuli are encoded in the normalized spiking activity of a population of tuned neurons (Bays, 2014), to provide a principled account of these stimulus-specific effects. While the original model assumed homogeneous tuning curves (Figure 1A), here we allow each neuron's tuning function to vary according to the principle of efficient coding (Figure 1B; Barlow, 1961; Ganguli & Simoncelli, 2014; Wei & Stocker, 2015). This principle states that neural responses should be optimized with respect to the natural frequency of stimuli in the environment. For orientation this means incorporating a prior that favors cardinal over oblique orientations. While continuing to capture changes to the mean distribution of errors with set size, the resulting model also reproduced stimulus-specific variations in precision (Figure 1C). Additionally, the efficient coding model predicted a repulsive bias away from the cardinal orientations: such a bias was observed in human response data. The model provides a general framework for describing stimulus-specific variations in VWM recall that can be readily extended to other feature dimensions.

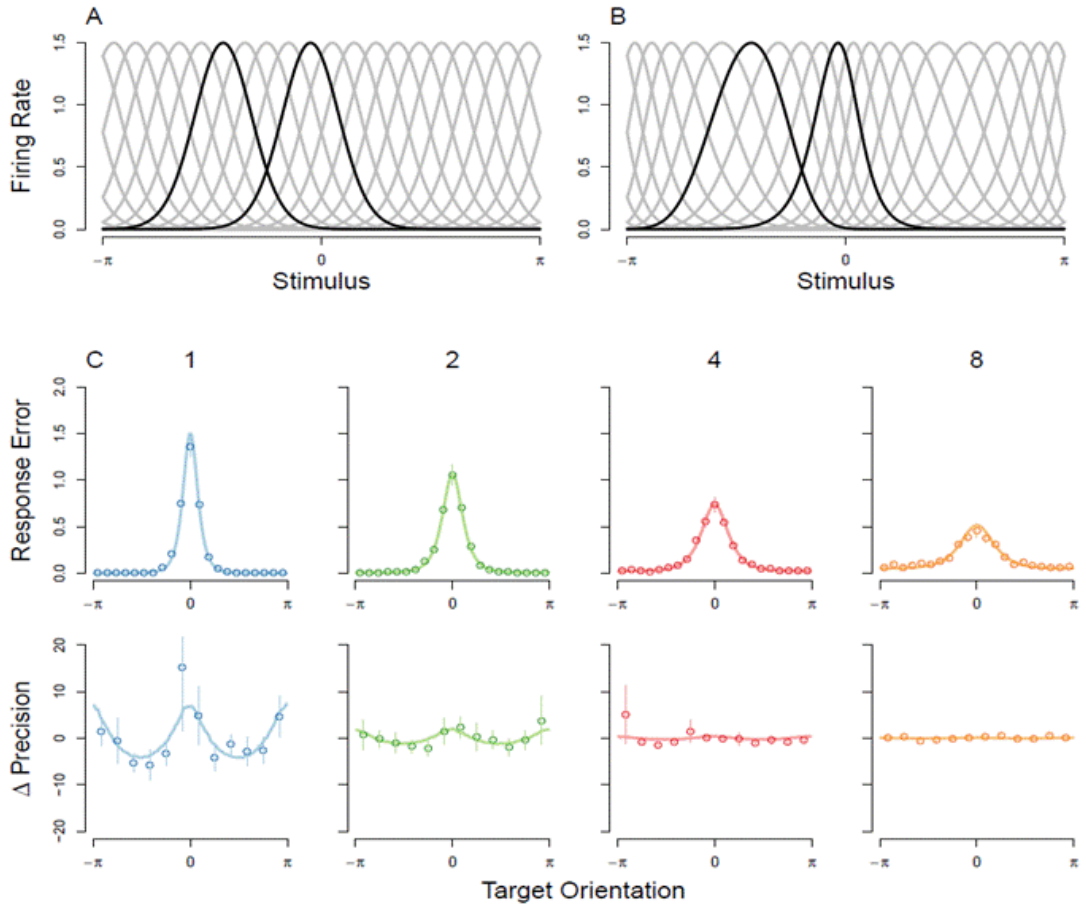


Figure 1. A) A homogenous population of Von Mises tuning curves. **B)** Under the principle of efficient coding, tuning functions become narrower, and more densely concentrated, around cardinal orientations (0, π). **C)** Distribution of recall errors for array sizes one to eight and precision across orientation space. Observers exhibit increased precision for cardinal orientations. Model fits shown as lines. Error bars represent 1± SE.

Disclosures: R. Taylor: None. **P.M. Bays:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.01/VV56

Topic: H.02. Human Cognition and Behavior

Title: What about grades in school?: the role of the working memory and fluid intelligence

Authors: *J. SIBAJA-MOLINA[1], O. A. RODRÍGUEZ-VILLAGRA[2,3]
[1]Neurosci. Res. Ctr., Univ. of Costa Rica, San Jose, Costa Rica; [2]Inst. for Psychological Res., [3]Neurosci. Res. Ctr., Univ. of Costa Rica, San José, Costa Rica

Abstract: Working Memory (WM) and Fluid Intelligence (FI) are described as two abilities associated with better adaptation in educational contexts and academic achievement. WM is a cognitive system related to temporary storage and manipulation of information for goal-directed processing. FI is defined as the ability to solve complex and novel problems, and formation of concepts, independent of learning and experience. In this behavioral study, we examined whether individual differences in WM and FI in preschoolers, predict the average grades in basic subjects (e.g., Math, Spanish, Science, Social Studies) at the first school year (i.e., academic achievement).We assessed WM and FI abilities of 132 students from public preschools located in San José, Costa Rica (~5.9 years; 67 girls; Spanish speakers).Two years after their preschool evaluation, grades were requested to the institutions where the children were enrolled in their first grade. Structural equation models were conducted to analyze the data. Tests of factorial invariance showed that all parameters of the measurement model were equivalent across girls and boys, thereby allowing the comparison of the mean structures across sex. Our main finding was that WM, but not FI, predicts the academic achievement factor. In the latter factor, a significant correlation between Spanish and Mathematics improved the fit of the model to the data suggesting further common aspects between these subjects. These results provide evidence of cognitive elements that underlie school grades. This information is relevant for educational curricula development and it shows the key role of WM in complex cognitive processes, which develop incrementally across early school stages. Understanding of WM is a hot topic in neuroscience research, and it is also important in fields like Education because it offers knowledge about the brain function that underlies some aspects of learning. Based on the behavioral results of this study, the current research in our laboratory is aimed at finding encephalographic correlates of WM (e.g. frontal midline theta rhythms) in early scholar ages, with the purpose to provide early measures that allow us to detect futures learning problems.

Disclosures: J. Sibaja-Molina: None. **O.A. Rodríguez-Villagra:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.02/VV57

Topic: H.02. Human Cognition and Behavior

Support: National Research Foundation of Korea (NRF-2016R1C1B2016039)

KAIST (Future Systems Healthcare Project)

Title: Pre-allocation of memory resources improves working memory performance in a sequential memory task

Authors: *H. LEE[1], Y. PARK[1], S.-B. PAIK[1,2]
[1]Bio and Brain Engin., [2]Program of Brain and Cognitive Engin., Korea Advanced Inst. of Sci. and Technol., Daejeon, Korea, Republic of

Abstract: Since working memory capacity is limited to a small number of items (Christophel et al., 2017), efficient use of memory resources is an important issue. Pre-allocation, the process of determining where memory will be stored before information comes in, is used in various memory systems, including computers, to utilize storage space. However, it is not clear whether pre-allocation of working memory exists in the brain. In this study, we propose that pre-allocation of working memory may exist, and it may be possible to improve memory performance based on human psychophysical experiments. We suggest a possible neural mechanism of pre-allocation using a neural network model.

First, to examine the effect of pre-allocation, we designed an experiment in which subjects memorized sequentially presented visual patterns under the conditions whether the subjects were (1) not informed or (2) correctly informed of the number of stimuli. For further analysis, (3) the subjects were informed of an incorrect number of stimuli to investigate how false information affects memory performance. Our results showed that performance for the early-presented items in the sequence was higher in the correctly informed case than in the other two cases. This suggests that memories for the early-presented items were less overwritten by recently-presented items when subjects were correctly informed, which indicates the presence of memory pre-allocation.

Next, to explain the systematic basis of experimental results, the concept of ‘neuronal ensemble’ (Josselyn et al., 2015) was applied. We designed a neural network model which learns input patterns and generates consistent output for trained patterns (Park et al., 2016). A group of neurons that selectively responded to learned inputs were defined as a ‘neuronal ensemble,’ and we could freely control the probability that a neuron would be recruited into the ensemble (pre-allocated) by modulating neuronal excitability (Silva et al., 2009). Using this model, we showed that the pre-allocation prevents old memories from being overwritten by new memories because it reduces the overlap between multiple ensembles. On the other hand, without pre-allocation, old memories were easily overwritten by new memories due to the large overlap between ensembles. These results are comparable with those obtained for the ‘correctly informed’ and ‘uninformed’ cases, respectively.

Overall, our results suggest that memory pre-allocation is in effect; thus, working memory performance is improved in the sequential presentation. The overlap of neuronal ensembles is one possible way to describe the effects of pre-allocation on memory performance.

Disclosures: H. Lee: None. **Y. Park:** None. **S. Paik:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.03/VV58

Topic: H.02. Human Cognition and Behavior

Support: NIH Grant R01 EY025278

Title: Neither cholinergic nor dopaminergic enhancement improves spatial working memory precision in humans

Authors: A. N. HAREWOOD SMITH[1], J. ADITYA CHALLA[2,3], *M. A. SILVER[4,1,3]
[1]Vision Sci. Grad. Group, [2]Dept. of Electrical Engin. and Computer Sci., [3]Helen Wills Neurosci. Inst., Univ. of California, Berkeley, Berkeley, CA; [4]Sch. Of Optometry, Univ. Of California, Berkeley, Berkeley, CA

Abstract: Acetylcholine and dopamine are neurotransmitters that play multiple important roles in perception and cognition. Pharmacological cholinergic enhancement reduces excitatory receptive field size of neurons in marmoset primary visual cortex, increases learning and performance of a spatial navigation task in rats, and sharpens the spatial tuning of visual perception in humans. Moreover, previous studies have shown that manipulation of dopaminergic signaling alters the spatial tuning of macaque prefrontal cortical neurons during the delay period of a spatial working memory task and can improve spatial working memory performance in human subjects. Here, we investigated the effects of systemic cholinergic and dopaminergic enhancement on a behavioral measure of the precision of spatial working memory in human subjects. Cholinergic transmission was increased by administration of 5 mg of the cholinesterase inhibitor donepezil, and dopaminergic signaling was enhanced with 100 mg L-dopa/10 mg carbidopa. Each neurotransmitter system was separately studied in double-blind placebo-controlled experiments. On each trial of the spatial working memory task, a square was presented for 150 ms at a random location along an invisible circle with a radius of 12 degrees of visual angle, followed by a 900 ms delay period with no stimulus shown on the screen. Then, the square was presented at a new location, displaced in either a clockwise or counterclockwise direction along the circle from the original location. Subjects compared their memory of the location of the original square to the location of the subsequent presentation and reported the direction of displacement. We defined spatial working memory precision as the amount of displacement corresponding to 75% correct performance. We observed no significant effect on spatial working memory precision for either donepezil or L-dopa/carbidopa. There was also no significant effect on performance on the spatial working memory task (percent correct across a wide range of displacements) for either donepezil or L-dopa/carbidopa. Thus, despite evidence that acetylcholine and dopamine can regulate spatial tuning of individual neurons,

pharmacological enhancement of signaling of these neurotransmitters does not substantially affect a behavioral measure of the precision of spatial working memory in humans.

Disclosures: **A.N. Harewood Smith:** None. **J. Aditya Challa:** None. **M.A. Silver:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.04/VV59

Topic: H.02. Human Cognition and Behavior

Support: Universidad de Guanajuato

Title: Electroencephalographic features during the performance of a test demanding working memory in early postmenopausal women

Authors: ***M. SOLIS-ORTIZ**[1], E. G. GONZALEZ-PEREZ[2], M. L. GUTIERREZ-MUÑOZ[3]
[1]Inst. Invest Med, Univ. Guanajuato, Leon, Mexico; [2]Med. Sci., Univ. de Guanajuato, Leon, Mexico; [3]Psicologia, Univ. Autonoma de Nuevo Leon, Monterrey, Mexico

Abstract: Studies examining the influence of the menopause on cognitive function have been contradictory. Working memory is key to successful cognitive processing, however how it co-varies with electroencephalographic (EEG) activity in early postmenopausal women is not well known. This study was aimed to explore the EEG features during the performance of a test demanding working memory based in a multivariate analysis. Twenty-five early postmenopausal healthy women between 48 and 60 years participated in the study. EEG was recorded during performance of a test demanding working memory. Absolute power of delta, theta, alpha1, alpha2, beta1, beta2 bands, the number of categories completed, correct responses (trials) as well as perseverative errors and errors of test were obtained and submitted to a principal components analysis. Four eigenvectors that accounted the 79.12 % of the total variance were identified. Theta, alpha1 and alpha2 bands were grouped together in an independent eigenvector (component 1), which explained 45.87 % of the total variance. The component 2 grouped beta2 band, and the component 3 grouped delta band, which explained 16.54 % and 11.49 % of the total variance, respectively. The component 4 grouped alpha2 band and the perseverative errors and errors of test in the occipital derivations, which explained 5.22 % of the total variance. The execution of the test was relatively poor. The number of categories achieved was less three out of a total number of six, and there were many errors. These findings show that during working memory processing theta and alpha bands, involved in activation of the working memory, executive performance and sustained attention, did not co-vary with beta2 and delta bands, which have been involved in alertness, focused arousal and inhibition of internal attention. The

errors committed covaried together with alplha2 band and they were independent of the other variables EEG.

Disclosures: M. Solis-Ortiz: None. **E.G. Gonzalez-Perez:** None. **M.L. Gutierrez-Muñoz:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.05/VV60

Topic: H.02. Human Cognition and Behavior

Support: JSPS KAKENHI JP16H02839

Title: Intersubject correlation analysis of brain activity when viewing videotaped teacher's explanation

Authors: *Y. HIRAKO[1], T. ITO[2], S. SHIMADA[3]
[1]Meiji Univ. Grad. Sch., Kawasaki-Shi, Japan; [2]Sch. of Arts and Letters, Meiji Univ., Ochanomizu, Japan; [3]Sch. of Sci. and technology, Meiji Univ., Kawasaki-Shi, Japan

Abstract: In recent years, educational methods using videotaped lessons have attracted attention. Previous studies showed that brain activities during viewing video movies have been studied using intersubject correlation (ISC) analysis. This study investigated the mechanism of comprehending videotaped teacher's explanation by utilizing ISC analysis of students' brain activity by using functional near-infrared spectroscopy (fNIRS). Twenty-four right handed subjects participated in the experiment (one female, aged 21.3 ± 0.17 years, mean ± SD). Half of the subjects (N=12) watched the explanation of a teacher about basic probability statistics in the recorded video. Another half watched a video clip of the equivalent explanation performed by another teacher. The hemodynamic responses in the bilateral cortical areas (9 x 9 square cm area each) were recorded by using 48-ch fNIRS. The sampling frequency was 10 Hz. The measured brain activity was analyzed by ISC analysis between two subjects who watched the same teacher (valid pairs) or different teachers (invalid pairs). ISC is calculated by utilizing general linear model (GLM) with one subject's brain activity as a model to see how similar the other subject's brain activity was. We also calculated dynamic ISC using 150 sample moving window (15.0s), on the channel that showed the significant difference in ISC between the groups. The ISC analysis showed a significant difference in ISCs between the valid pairs and the invalid pairs in the right DLPFC (ch-38: $t(24) = 1.78$, $p < 0.05$). We investigated the time series change of the ISC in ch-38 and found that they were consecutively significant ($z > 1.66$, $p < 0.05$) in the scenes that the teacher was explaining "variance" (teacher A : 120.0s - 136.4s, teacher B : 156.3s - 177.4s) and "standard deviation" (A : 176.0s - 207.0s, B : 202.5s - 226.6s), where the

explanation is relatively easy to understand for subjects as the teacher read aloud sentences written in the slide. These results suggest that rDLPFC is involved in comprehending videotaped teacher's explanation, possibly by playing a role as working memory to translate verbal information into abstract numerical representation.

Disclosures: **Y. Hirako:** None. **T. Ito:** None. **S. Shimada:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.06/VV61

Topic: H.02. Human Cognition and Behavior

Support: NIH Grant 5R01-MH087214-08

ONR N00014-12-1-0972

Title: Decoding the limits of simultaneous storage in working memory

Authors: *K. C. ADAM, E. AWH, E. K. VOGEL
Psychology, Univ. of Chicago, Chicago, IL

Abstract: Recent work has shown that the topography of power in the alpha-band (8-12 Hz) of the human EEG signal tracks the location of a single item held in visual working memory (Foster et al., 2016). A critical question in working memory (WM) research is the existence and nature of capacity limits. Most models of visual WM endorse a capacity limit of 3 simple items, but others have suggested that active representation may be limited to a single prioritized item. To best address capacity debates, we need methods for simultaneously decoding the identity of all items purportedly stored in WM. Here, we made advances toward this aim by presenting and decoding the identity of up to 3 simultaneously-presented items. One weakness of prior research in this area is the confound of increasing visual stimulation with an increasing number of items; this confound could lead to inadequate decoding for higher set-sizes because of increased noise at encoding. To control for visual stimulation between the different load conditions, we used a selective encoding working memory task. On every trial, three colored squares appeared in three of eight possible location bins equidistant from fixation. To manipulate memory load, participants were pre-cued to 1, 2, or 3 of the relevant locations with centrally presented spatial cues (small lines pointing toward the location or locations). We found reliable decoding of all 3 load conditions using alpha-band activity. The selectivity of decoding was highest for set-size 1 trials, and then declined for set-sizes 2 and 3. Our findings are consistent with previous work demonstrating a decrement in behavioral precision for increased memory loads, and demonstrate the feasibility of extending alpha-based decoding to test the limits of capacity.

Disclosures: **K.C. Adam:** None. **E. Awh:** None. **E.K. Vogel:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.07/VV62

Topic: H.02. Human Cognition and Behavior

Support: NIMH Grant 2R01-MH087214-06A1

Title: Alpha-band activity reveals concurrent storage of independent locations in spatial working memory

Authors: ***J. J. FOSTER**, D. W. SUTTERER, K. C. ADAM, E. K. VOGEL, E. AWH
Univ. of Chicago, Chicago, IL

Abstract: Past work has shown that the scalp distribution of oscillatory alpha-band (8-12 Hz) activity - measured with electroencephalography (EEG) - precisely tracks locations held in working memory (WM). Thus, spatially selective tuning profiles (called channel-tuning functions or CTFs) can be reconstructed from the spatial distribution of alpha power using an encoding model of spatial selectivity (Foster et al., 2016). Recent work using functional magnetic resonance imaging (fMRI) has shown that feature-selective patterns of BOLD activity degrade as the number of items to be stored increases (e.g., Sprague et al., 2014). Here, we examined whether increased storage loads elicit a similar decline in the spatial selectivity of alpha-band activity. On each trial, subjects encoded and maintained the location of one or two colored dots while we recorded EEG. After a 1s delay period, subjects were cued to report the location of one of the dots. The set size 2 condition revealed concurrent alpha-band CTFs that tracked the position of each item throughout the delay period, but with reduced spatial selectivity relative to the set size 1 condition. In addition, set size 2 data was robustly decoded using a set size 1 training set, showing that subjects did not rely on a configural code to accomplish simultaneous storage of the items in the set size 2 condition. Likewise, simulations revealed that CTF selectivity in the set size 2 condition was high enough to rule out trial-by-trial switching of attention between the items in the set size 2 condition, bolstering evidence for concurrent storage of independent feature values in working memory. Thus, oscillatory activity in the alpha band enables time-resolved tracking of concurrent representations in visual WM and dovetails with past observations that mnemonic quality declines as storage load increases.

Disclosures: **J.J. Foster:** None. **D.W. Sutterer:** None. **K.C. Adam:** None. **E.K. Vogel:** None. **E. Awh:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.08/VV63

Topic: H.02. Human Cognition and Behavior

Support: NIH/NIMH Grant 5R01MH087214

Title: Evidence for concurrent activation of sequentially encoded spatial locations

Authors: *D. W. SUTTERER[1,2], E. AWH[2]
[2]Psychology, [1]Univ. of Chicago, Chicago, IL

Abstract: A robust body of work has demonstrated speeded reaction times to the last item encoded in a Sternberg task (Vergauwe et al., 2016, Oztekin, Davachi, & McElree 2010). A key debate is whether this RT benefit reflects a narrow focus of attention on the most recently presented item, in line with suggestions that working memory capacity may be limited to a single item or encoding episode. Here, we tested this hypothesis by using alpha band activity to track the time course and content of representations maintained in spatial working memory (Foster et al., 2016). This method allowed us to observe whether or not sequentially encoded items are concurrently represented during the delay period of a working memory task. On each trial, observers memorized the location of two sequentially presented colored dots while EEG was recorded. After a 1s delay period, participants were cued to report the location of one of the dots. We trained an inverted encoding model (IEM) to assess alpha selectivity for each item and found robust representations of both to-be-remembered items during the 1s delay interval consistent with the interpretation that both items are simultaneously maintained in an active state. These findings disconfirm the hypothesis that only the final item in a sequence is actively represented during the delay period. Thus, although faster RT for the final item in a sequence may reflect a higher priority or familiarity for the most recently encoded item, multiple items in a sequence can be simultaneously stored in visual working memory.

Disclosures: D.W. Sutterer: None. **E. Awh:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.09/VV64

Topic: H.02. Human Cognition and Behavior

Support: Wellcome Trust Grant 106926

Title: Restoration of fMRI decoding quality does not imply latent working memory states: A neural field model of retro-cue effects

Authors: *S. SCHNEEGANS, P. M. BAYS
Dept. of Psychology, Univ. of Cambridge, Cambridge, United Kingdom

Abstract: The prevailing view of working memory is that memory representations are maintained through sustained spiking activity in neural populations. This view has recently been challenged by studies using machine learning methods to decode memory content from human fMRI and EEG data (Sprague et al., 2016; Rose et al., 2016; Wolff et al., 2017). These studies found that previously diminished decoding fidelity of an item was restored following an informative cue or non-specific stimulation. This was interpreted as evidence for an activity silent working memory state (realized e.g. through synaptic changes), which can itself not be decoded from neural activity, but which can be restored into an active state or modulate activity patterns in response to external stimulation. Here, we test the validity of this conclusion by formulating a neural network model that employs sustained neural activity as its sole memory mechanism, and represents feature combinations through conjunctive coding. We apply this model to a spatial recall task with retrospective color cueing as used in a recent fMRI study, and demonstrate that it can reproduce both behavioral and fMRI decoding results. In particular, an informative color cue in the model strengthens the spatial representation of the cued item and suppresses memory representations of non-cued items within the conjunctive space-color map. This produces an increase of decoding quality following the color cue (Figure 1), replicating the key effect of the fMRI study that was claimed as evidence for latent working memory. These results demonstrate that activity-silent memory states are not necessary to explain restoration of decoding quality, and highlight how sustained activity in a neural system can interact in complex ways with new inputs.

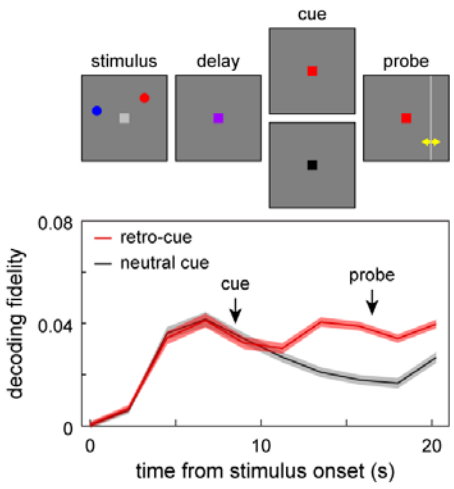


Figure 1: Psychophysical task and decoding fidelity for the target location in the neural network model. Fidelity is computed from simulated BOLD signals based on the activity in the neural network, emulating the reconstruction approach of a recent fMRI study (Sprague et al., 2016). In the task, subjects were presented with two colored disks, and after a delay had to report one location cued by the corresponding color. In the absence of an informative cue, decoding fidelity decays over the course of the delay period. But following a color retro-cue that indicates which item will be tested, decoding fidelity for the target location recovers significantly. An analogous finding in the fMRI data has been interpreted as evidence for a latent working memory state, but in the neural model it is produced by interactions between sustained memory activity and the color cue.

Disclosures: S. Schneegans: None. **P.M. Bays:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.10/VV65

Topic: H.02. Human Cognition and Behavior

Title: Associations between MEG-measured prefrontal activity during working memory and dopamine D_1 receptor availability measured with PET

Authors: *D. Y. RUBINSTEIN[1], D. P. EISENBERG[1], A. M. IANNI[1], F. W. CARVER[2], D. R. WEINBERGER[3], R. COPPOLA[2], K. F. BERMAN[1]
[1]Section on Integrative Neuroimaging Clin. & Translational Neurosci. Br., [2]MEG Core Facility, Natl. Inst. of Mental Hlth., Bethesda, MD; [3]Lieber Inst. For Brain Develop., Baltimore, MD

Abstract: A relationship between working memory-related dorsolateral prefrontal cortical (DLPFC) activity and dopamine D_1 receptor signaling within the prefrontal cortex has been well studied in non-human primates, but it has been difficult for human studies to achieve similar specificity in the measurement of the electrophysiological and neurochemical components underlying this relationship. To better elucidate the association between DLPFC activity and D_1 receptor biology in healthy volunteers, we used magnetoencephalography (MEG) to measure neuronal activity and PET to measure DLPFC-specific D_1 receptor function and tested the relation between the two parameters. In a large sample of healthy volunteers (N=342), neuromagnetic activity was recorded with a 275 channel whole head MEG during performance of 2-back working memory and 0-back sensorimotor control conditions. Source localization was achieved with an adaptive beamformer, which was used to estimate the 2-back to 0-back power ratio at 5mm-spaced points throughout the brain in 400ms windows time-locked to correct responses, in theta, alpha, beta, and gamma frequency bands. In a subsample of this cohort (n=40), [^{11}C]NNC112 PET scanning was also performed, beginning with an 8-minute transmission scan to correct for attenuation, followed by a 90-minute emission scan. Receptor availability was assessed by calculating the non-displaceable binding potential (BP_{ND}) at each voxel in a subject's brain, using PMOD to implement a simplified reference tissue model with a cerebellar reference region. An a priori, cytoarchitectonically-based mask of the DLPFC from the literature was used to extract MEG and PET values for correlative analysis. While all frequency bands exhibited various robust working memory-related power modulations across the brain, only beta band revealed a fronto-parietal activation pattern similar to that observed with other modalities. In the context of this activation pattern, we found robust working memory-related beta band DLPFC desynchronization (activation) in the time window centered on 200ms before correct responses ($p < .001$). Within the subsample who also had PET, individuals with greater DLPFC activation also exhibited higher D_1 receptor availability in the same region ($r = .42$, $p < .01$), after adjusting for effects of age. These results are consistent with primate studies

demonstrating an important role for D_1 receptors in the modulation of prefrontal WM-related activity, and are among the most direct evidence of such a relationship in humans. Future research in patients will help determine dopaminergic mechanisms underlying their prefrontal, WM-related deficits.

Disclosures: D.Y. Rubinstein: None. **D.P. Eisenberg:** None. **A.M. Ianni:** None. **F.W. Carver:** None. **D.R. Weinberger:** None. **R. Coppola:** None. **K.F. Berman:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.11/VV66

Topic: H.02. Human Cognition and Behavior

Support: NSF GRFP

CIFAR

Title: A self-organizing memory network

Authors: *C. FEDERER[1], J. ZYLBERBERG[2]
[2]Physiol. and Biophysics, [1]Univ. of Colorado Sch. of Med., Aurora, CO

Abstract: Working memory relies on us being able to retain information about stimuli even after they go away. Stimulus information is encoded in the activities of neurons, which change over timescales of milliseconds[1]. Information in working memory can be retained for tens of seconds, leaving the question of how time-varying neural activities keep representing the same information. Prior work shows that, if the neural dynamics are in the “null space” of the representation - so that changes to neural activity do not affect the downstream read-out of stimulus information - then information can be retained for periods much longer than the time-scale of individual-neuronal activities[2]. The prior work, however, has a fine-tuning problem: precisely constructed synaptic connectivity matrices are needed for information to be retained, and it is unclear how biological circuits would obtain that level of precision. To identify mechanisms through which biological networks can self-organize to support memory function, we derived biologically plausible synaptic plasticity rules that dynamically organize the connectivity matrix to enable retention of stimulus information. Networks implementing this plasticity rule can successfully information about stimulus even if only 10% of the synapses are plastic; they are robust to synaptic noise and can store information about multiple stimuli. Networks can also maintain information about stimulus even if only 10% of the network is connected. Networks that have been pre-trained on stimuli can continue to store new stimuli without being able to update the synapses. The derived synaptic updates require a global error

signal across the network. We propose two possible sources of this signal: calculations via segregated dendrites or from neuromodulators[3]. Our results suggest a biologically plausible model of stimulus retention with time varying neural activity and connectivity. **References** [1] Brody *et al*. (2003). *Cereb. Cortex.* [2] Druckmann & Chklovskii (2012). *Curr. Biol.* [3] Guergiuev *et al*. (2016) *arXiv preprint.*

Disclosures: C. Federer: None. **J. Zylberberg:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.12/DP14/VV67 (Dynamic Poster)

Topic: H.02. Human Cognition and Behavior

Support: NIH Grant F32 MH111204

Title: Network competition and reconfiguration during working memory processing

Authors: *A. KIYONAGA, D. J. LURIE, M. D'ESPOSITO
Helen Wills Neurosci. Inst., Univ. of California Berkeley, Berkeley, CA

Abstract: A core function of working memory (WM) is to temporarily maintain information in the face of disruptions and more immediate attention-demanding tasks. It is unclear, however, why simultaneous processing demands can have widely variable degrees of impact on WM performance, and how the brain manages the competition between demands to maximize performance. Prefrontal and parietal brain regions have long been implicated in WM maintenance and dual-task performance, but we lack a firm grasp of their specific contributions to these processes. Here, we examine interactions between these brain regions, and their roles in large scale brain network organization—during several types of WM disruption—to achieve a clearer picture of the neural circuitry of WM dual-task processing. During fMRI scanning, participants completed a visual WM delayed match-to-sample task with an intervening visual search demand embedded in the delay. Delay-spanning processing demands were factorially manipulated on two levels: 1) interference from perceptually dissimilar or similar visual search stimuli and 2) attentional demand from an easier or harder visual search task. Behavior was impacted by both interference and attentional demand, as well as an interaction between factors. WM recognition performance was worst when demands were high on both factors. This interaction was also reflected in univariate BOLD activity in prefrontal and parietal regions that are typically engaged when task demands are high. Specifically, when dual-task demands were greatest, activity peaked in the rostral portions of prefrontal cortex that have been implicated in the coordination of multiple simultaneous demands. Moreover, separate “cognitive localizer” tasks identified distinct but overlapping networks that were engaged by WM vs. attentional

orienting demands, and a greater extent of overlap between these networks related to impaired dual-task performance across individuals, suggesting that better segregation between functional networks can facilitate efficient dual-task processing. These data illustrate how competition emerges between task demands at the whole brain network level, and shed light on how task-based functional network configuration can protect WM content from disruption.

Disclosures: A. Kiyonaga: None. **D.J. Lurie:** None. **M. D'Esposito:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.13/VV68

Topic: H.02. Human Cognition and Behavior

Support: NIH R01-EY022229

NIH F31-MH101963

NIH F32-EY026796

CELEST an NSF Science of Learning Center SMA-0835976.

Title: Disentangling sensory specialization from task specialization in lateral frontal cortex

Authors: *A. L. NOYCE[1], N. CESTERO[2], S. W. MICHALKA[3], B. G. SHINN-CUNNINGHAM[2], D. C. SOMERS[1]
[1]Psychological and Brain Sci., [2]Boston Univ., Boston, MA; [3]Olin Col. of Engin., Wellesley, MA

Abstract: Prior work has suggested that much of human lateral frontal cortex (LFC) is part of a 'multiple demand' network, supporting a wide range of cognitive tasks and processes (e.g. Duncan 2010; Fedorenko et al. 2013). In contrast to this multiple-demand view, several groups have recently reported that portions of LFC show a preference for processes associated with a particular sensory modality (Michalka et al. 2015; Braga et al. 2016; Mayer et al. 2016). Michalka et al. demonstrated that selective spatial attention to visual or auditory stimuli recruits specific structures within LFC. Two bilateral regions are biased for visual attention, superior and inferior precentral sulcus (sPCS and iPCS), and two bilateral regions are biased for auditory attention, transverse gyrus intersecting precentral sulcus (tgPCS) and caudal inferior frontal sulcus (cIFS). Here, we used fMRI to replicate this finding in a substantially different task paradigm and to examine the 'multiple demand' responsiveness of these regions. We observed that visual and auditory 2-back working memory tasks recruited interleaved visual- and auditory-biased structures that correspond to those reported by Michalka et al. These structures were mapped in each individual subject (n = 15). Within-subject reliability (n = 7) was high across the

two tasks. We then tested the degree to which these sensory-biased structures demonstrated multiple demand behavior. First, we measured BOLD recruitment in the non-preferred memory task; visual-biased sPCS and iPCS showed significantly more recruitment during auditory WM than did tgPCS and cIFS during visual WM. Second, for each vertex within these structures we projected the activation in the two tasks into a 2D vector space; the multiple demand index derived from these values was significant for visual-biased but not for auditory-biased LFC structures. Finally, MVPA stimulus classification tested the degree to which the structures carried information about two classes of visual and two classes of auditory stimuli; visual-biased sPCS and iPCS carried high amounts of information about both stimuli sets, but auditory-biased tgPCS and cIFS carried significantly less information about visual than about auditory stimuli. All three metrics indicate that the visual-biased LFC structures exhibit stronger multiple-demand responsiveness than do the auditory-biased structures, suggesting that the auditory processing network may be more specialized in its cognitive role. These results reconcile two competing theories of LFC's contribution to human cognition by demonstrating the coexistence of sensory specialization and multiple demand behavior.

Disclosures: **A.L. Noyce:** None. **N. Cestero:** None. **S.W. Michalka:** None. **B.G. Shinn-Cunningham:** None. **D.C. Somers:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.14/VV69

Topic: H.02. Human Cognition and Behavior

Support: R01-EY016407

Title: Spatially specific delay period activity in the human superior colliculus

Authors: *K. DESIMONE[1,4], M. RAHMATI[2], G. T. SABER[3], K. K. SREENIVASAN[4], C. E. CURTIS[5]
[1]Dept. of Psychology, [2]New York Univ., New York, NY; [3]New York Univ., Charleston, NY; [4]New York Univ. Abu Dhabi, Abu Dhabi, United Arab Emirates; [5]Psych & CNS, NYU, New York, NY

Abstract: The superior colliculus (SC) is a key node in a distributed oculomotor network and mediates orienting behaviors such as saccadic eye-movements and gaze shifts. The SC is a laminar structure sitting atop the brainstem and contains two tightly registered retinotopic maps: a visual map in the superficial layer, and a motor map in the intermediate and deep layers representing the angle and amplitude of saccades (Wurtz & Albano, 1980; Sparks, 1986). However, this classical view of the response properties of the SC has been challenged by

pharmacological inactivation of the deep layers of the SC in the macaque, which induces a form of visual neglect akin to extinction not accounted for by paresis or anopsia (McPeek & Keller, 2004; Lovejoy & Krauzlis, 2010). We sought to examine the extent to which the human SC is able to maintain representations of behavioral goals beyond simpler visual and/or motor responses. We hypothesized that the SC acts as a topographic map of spatial priority (Fecteau & Munoz, 2006), and predicted that we should find spatially specific representations of behavioral goals in human SC during a delay period between visual stimulation and motor execution. To test this, we used fMRI to measure SC activity while participants performed memory-guided pro- and anti-saccades. The anti-saccade trials provide the key test of our hypothesis, as these trials dislocate visual and motor responses in space. We examined the trial-wise time series of the population activity in the SC during the anti-saccade task. In addition, we used an inverted encoding model (Sprague & Serences, 2013) to characterize the topographic organization of SC activity during the task. The activity of each SC voxel was modeled as the weighted sum of a set of spatial basis functions spanning the locations of visual stimuli and saccade targets. These channel weights were then combined across voxels to generate reconstructions that recast the SC population activity during pro- and anti-saccade trials into visuotopic coordinates. First, we found robust and spatially specific univariate persistent activity during the delay period following presentation of the visual cue and but prior to execution of the saccade. This suggests that populations of neurons in the human SC are able to maintain task-relevant information throughout a long delay. Second, we also found that the early delay period activity of the SC reflected the location of the visual stimulus. However, as the delay period progressed, the locus of activity shifted from the visual stimulus to the location of the saccade endpoint. Thus, the dynamics of the human SC tracked the spatial and temporal aspects of the behavioral goal.

Disclosures: K. DeSimone: None. **M. Rahmati:** None. **G.T. Saber:** None. **K.K. Sreenivasan:** None. **C.E. Curtis:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.15/VV70

Topic: H.02. Human Cognition and Behavior

Support: NIMH IRP

Title: Association of hippocampal D_1/D_2 receptor availability and neural activity during implicit encoding

Authors: *S. E. GROGANS[1], R. RASETTI[2], M. GREGORY[1], B. KOLACHANA[1], M. WINSTON[1], C. HEGARTY[1], A. IANNI[1], P. KOHN[1], J. H. CALLICOTT, III[3], D. R. WEINBERGER[4], K. F. BERMAN[5]

[1]Natl. Inst. of Mental Hlth., Bethesda, MD; [2]CBDB, NIMH, NIH, Bethesda, MD; [3]DIRP, NIMH, NIH, Clin. and Translational Neurosci. Br., Bethesda, MD; [4]Lieber Inst. For Brain Develop., Baltimore, MD; [5]Section on Integrative Neuroimaging, Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: Hippocampal dysfunction is implicated in several neuropsychiatric conditions, including Alzheimer's disease and schizophrenia. The hippocampus (HIP) receives important dopaminergic input from the VTA and contains populations of both D_1 and D_2 receptors, which may be relevant to this structure's function in a way that could inform disease models. Prior research has suggested mnemonic functions may be reliant on HIP recruitment and HIP availability of dopamine receptors; additionally, activation of D_1 receptors alter excitability in the HIP and therefore influence memory encoding. However, work in other regions of the cortex propose that D_1/D_2 ratios determine signal-to-noise during cognitive processing. Based on these previous data, we hypothesized that increased HIP D_1/D_2 availability ratio would be positively correlated with increased HIP activation during episodic memory in a sample of healthy volunteers.

Fifty-four healthy volunteers (mean age=33.8±9.4; 24 women) underwent both PET and fMRI investigations. PET imaging measured DA receptor availability with [^{11}C]-NNC 112 (D_1 receptors) and [^{18}F]-fallypride (D_2 receptors), using time activity curves from the cerebellum and bilateral HIP as reference and of-interest regions, respectively, and non-invasive simplified reference tissue modeling. FMRI measured BOLD activation during the neutral image encoding condition of a simple implicit memory task well known to engage the HIP. We tested for voxelwise correlations between BOLD activation and the ratio of HIP binding potentials (BP_{ND}; D_1/D_2 ratio) for [^{11}C]-NNC 112 and [^{18}F]-fallypride. Results were considered significant at $p<0.05$, FWE-corrected within the HIP region-of-interest. Whole-brain analyses and retrieval performance effects were also examined.

HIP D_1/D_2 ratio (but not D_1 nor D_2 availability individually) positively correlated with activation in the left HIP ($p_{FWE\text{-}corrected\text{-}within\text{-}ROI}=0.009$). In the whole-brain analysis, hippocampal D_1/D_2 ratio was positively correlated with activation in dorsal anterior cingulate ($p_{FWE\text{-}corrected\text{-}whole\text{-}brain}=0.015$). Neither of these results changed when age and gender were used as covariates of no interest. Additionally, HIP D_1/D_2 ratio was not associated with performance.

Our results suggest that relative D_1/D_2 receptor availability in HIP may have importance in the neurophysiology of novel stimuli visual encoding. These findings provide insight into the role of DA receptors in episodic memory, suggesting that relatively increased D_1 with respect to D_2 may be advantageously associated with increased HIP engagement.

Disclosures: S.E. Grogans: None. **R. Rasetti:** None. **M. Gregory:** None. **B. Kolachana:** None. **M. Winston:** None. **C. Hegarty:** None. **A. Ianni:** None. **P. Kohn:** None. **J.H. Callicott:** None. **D.R. Weinberger:** None. **K.F. Berman:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.16/VV71

Topic: H.02. Human Cognition and Behavior

Support: ONR MURI N00014-16-1-2832

NIH Grant R01 MH111737

Title: Neurodynamic mechanisms of working memory gating

Authors: *B. J. FRICK[1], C. W. HOY[2], J. LIN[4], R. T. KNIGHT[5,3,6], M. D'ESPOSITO[5], D. BADRE[1]
[1]CLPS, Brown Univ., Providence, RI; [2]Helen Wills Neurosci. Inst., [3]The Psychology Dept., Univ. of California at Berkeley, Berkeley, CA; [4]Comprehensive Epilepsy Program, Dept. of Neurol., Univ. of California, Irvine, Irvine, CA; [5]Helen Wills Neurosci. Inst., Univ. of California Berkeley, Berkeley, CA; [6]Dept. of Neurolog. Surgery and Physiology,, Univ. of California, San Francisco, CA

Abstract: Cognitive control requires working memory (WM) to maintain relevant information that provides a context for behavior. Efficient use of WM resources requires selectively encoding new representations (input gating) and selecting stored information to exert a contextual influence (output gating). In EEG, MEG, and LFP studies, several power-spectral features have been identified as mechanistic markers for exertion of cognitive control in frontal cortex. Specifically, intra-cortical beta events, frontal theta-synchrony, and PFC beta-gamma and theta-gamma interactions have been implicated with WM related functions. These functions are likely implemented by timing-dependent mechanisms in the frontal cortex. Within hierarchical cognitive control, the ordering of contextual information may influence these neural mechanisms for the purpose of facilitating WM input and output gating. Prior work suggests that a largely overlapping corticostriatal network facilitates both gating mechanisms. However, the relationship of these gating mechanisms and oscillatory markers of cognitive control function remains unknown. In order to discover the neurodynamic functions that underlie the input/output gating of WM, we recorded from intracranial electrodes while human patients performed a WM gating task. In each trial, subjects were presented with either a *context-item-item* sequence, permitting input gating, or an *item-item-context* sequence, necessitating output gating, and were asked to recall which presented *item* corresponded to the presented *context*. We analyzed the power-spectral profiles of each event and found (1) differential theta activity during output gating, (2) beta burst activity for items informed by a input-gated context, and (3) temporally-specific interactions between regions and frequency bands across the frontal cortex. Together,

these results inform a neurodynamic model for input gating and output gating mechanisms within frontal cognitive control networks.

Disclosures: **B.J. Frick:** None. **C.W. Hoy:** None. **J. Lin:** None. **R.T. Knight:** None. **M. D'Esposito:** None. **D. Badre:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.17/VV72

Topic: H.02. Human Cognition and Behavior

Support: NSF BCS1554105

NIH R01MH110831

McKnight Endowment Fund for Neuroscience

NARSAD

Title: Load-related theta decrease in DLPFC during information maintenance in working memory predicts subsequent performance

Authors: *A. BRZEZICKA[1,3], J. KAMINSKI[1,4], A. MAMELAK[1], U. RUTISHAUSER[1,4,2]
[2]Dept. of Neurol., [1]Cedars-Sinai Med. Ctr., Los Angeles, CA; [3]SWPS Univ. of Social Sci. and Humanities, Warsaw, Poland; [4]Caltech, Pasadena, CA

Abstract: Theta (3-8 Hz) oscillatory activity is thought to be crucial for maintaining information in working memory (WM). Whereas in animal work, the focus is on hippocampal theta, human WM studies are focused on cortical frontal-midline theta (Mitchell et al., 2008). Results from both lines of work show theta power increase with memory load. Here, we focused on the relationship between load-related theta power changes in dorsolateral prefrontal cortex (DLPFC). DLPFC is thought to be critical for the maintenance of information in working memory as shown by many monkey single unit and human fMRI studies. We performed human intracranial recordings in 12 neurosurgical, epileptic patients. We recorded local field potential (LFP) signals from depth electrodes implanted in DLPFC during a modified Sternberg task with three levels of memory load and pictures as a study material. We localized the most lateral DLPFC contacts for each patient (total of 48 DLPFC electrodes). We found a surprising pattern of theta power changes: it increased in the beginning of the task compared to baseline (t_{48}=2.38, p=0.021; M_{bas} = 0.021 [SEM=0.008]; M_{task} = 1.64, [SEM=0.056]) but showed a substantial load-depended decrease during the maintenance period of the task ($F_{2,46}$=27.34; p<0.0001, eta^2=0.54, linear trend $F_{1,47}$=53.81, p<0.0001, eta^2=0.53). This load-dependent decrease in theta predicted

individual differences in performance of the Sternberg task. Such that the bigger the theta change the faster (r=-0.393, p=0.006) and more accurate (r=-0.289, p=0.046) was the subject. In contrast, in the hippocampus (24 electrodes in total) theta power increases as a function of memory load $_{F2,22}$ = 4,01, p=0.033, eta^2=0.267. Those striking differences suggest that cortical theta in the DLPFC reflects neuronal processes distinct from those reflected by hippocampal theta. Lisman and Jensen (2013) postulated that a signature of excited (or engaged in task) cortex is a reduction in the power of theta (similar to alpha band), so we hypothesize that decreases in power are related to more effortful task completion. Such thinking is also supported by previously shown negative relationships between cortical theta power and fMRI BOLD changes. In addition, recent work shows that the extent of theta power decreases in cortical areas, including the DLPFC, predicts how well information will later be remembered in long-term memory tasks (Greenberg et al., 2015). Our observations extend this phenomenon to DLPFC theta during maintenance of information in working memory.

References:
Greenberg et al. (2015). Neuroimage, 114.
Lisman, Jensen, (2013). *Neuron*, *77*(6).
Mitchell, et al. (2008). *Progress in neurobiology*, *86*(3).

Disclosures: **A. Brzezicka:** None. **J. Kaminski:** None. **A. Mamelak:** None. **U. Rutishauser:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.18/VV73

Topic: H.02. Human Cognition and Behavior

Title: The limits of unconscious working memory

Authors: ***M. PAYTON**[1], I. UL-HAQ[2], V. BABUSHKIN[4], K. K. SREENIVASAN[3]
[1]Psychology, New York Univ., New York, NY; [2]Div. of Sci. and Mathematics, [3]New York Univ. Abu Dhabi, Abu Dhabi, United Arab Emirates; [4]Div. of Sci. and Mathematics, New York Univ. - Abu Dhabi, Abu Dhabi, United Arab Emirates

Abstract: Working Memory (WM) is canonically defined as a conscious and effortful process that allows for the retention and manipulation of information over brief periods of time. Recent work has suggested that short-term maintenance can be achieved in the absence of subjective awareness of the memory item, challenging the long-held notion that WM is by definition a conscious process. In light of this work, an important goal is to understand which aspects of WM require conscious awareness.
The present study sought to test key properties of unconscious WM. Specifically, we were

interested in (i) whether unconscious WM representations can be manipulated; (ii) the degree to which features of an object can be bound together in unconscious WM; and (iii) the capacity limitations of unconscious WM. To address these questions, we had observers perform a delayed estimation task for spatial location. On each trial, observers were presented with a dot that appeared at a random location around an invisible annulus. The observers' task was to maintain the spatial location of this dot over a brief delay, and then report the remembered location with a mouse click. We manipulated conscious awareness of the dot stimulus using backward masking. At the end of each trial, observers rated their awareness of the dot stimulus on a standard 4-point perceptual awareness scale, a score of 4 indicating 'a very clear image' and a score of 1 indicating 'no information' (Sandberg et al., 2009). Analysis of unconscious data was limited to masked trials scored as 1 by the observer. To test whether unconscious WM representations could be manipulated, Experiment 1 required observers to mirror the remembered location over a randomly oriented line and subsequently rotate the mirrored location around the circle by 90 degrees. Subjects reported the end result of these two transformations. To test feature-binding and capacity limitations, Experiment 2 required observers to remember the locations of multiple colored dots and bind the dots' location and color.
Contrary to earlier studies on unconscious serial processing (Sackur & Dehaene 2009), Experiment 1 demonstrated that observers could perform serial spatial manipulations on unconscious WM representations. In Experiment 2, we found that observers could successfully perform feature binding at a set size of 2, although results were more variable as set size increased. Together, these results show that unconscious WM shares critical features with conscious working memory but with potentially stricter resource limitations.

Disclosures: M. Payton: None. **I. Ul-Haq:** None. **V. Babushkin:** None. **K.K. Sreenivasan:** None.

Poster

433. Working Memory

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 433.19/VV74

Topic: G.07. Other Psychiatric Disorders

Title: Ear morphometrics correlate with schizotypal inventory scores and SAT performance in sex- and side-specific manners

Authors: *J. CANNON[1], P. T. ORR[2]
[1]Dept. of Psychology, [2]Psychology Dept., Univ. of Scranton, Scranton, PA

Abstract: Our lab has been studying the relationships among facial features and a number of cognitive (e.g., SAT) and personality (e.g., schizotypal) characteristics (Cannon et al., 2015). Embryological development of the face and anterior brain occur in synchrony, are often

influenced by common genes, and have the potential to be dramatically and characteristically altered by genetic abnormalities or environmental insults. Adhikari et al. (2014) performed a genome-wide association study that identified multiple loci associated with variations in human ear morphology. They found moderate and significant heritability in all ear characteristics measured. Here we examined relationships among Adhikari et al.'s ear morphology features and our behavioral measurements.

Left and right digital profile photographs were taken of undergraduates (N = 110; 74.5% female). Scoring with the same 0-2 ranking system utilized by Adhikari et al., paired blind observers evaluated 9 of 10 ear characteristics examined by Adhikari et al. The 10th feature, Protrusion, was not interpretable in our profile images. Inter-rater correlations were high ($r > .84$) and averages of the paired observers were used for statistical analyses. Pearson correlations were performed among ear measurements and overall/subscale scores of the Schizotypal Personality Questionnaire (SPQ) and Math/Verbal SAT scores.

Significant for Averaged Ears in Males: Tragus Size (TS) with Verbal-SAT (-.564). Significant for Averaged Ears in Females: Crus Helix Expression (CH) with Cognitive/Perceptual SPQ factor (C/P) (-.268), CH with Ideas of Reference (IR) (-.308), Lobe Attachment (LA) with Odd/Eccentric Behavior (O/E) (-.251), Lobe Size (LS) with O/E (.309), and Helix Rolling (HR) with Constricted Affect (CA) (.372).

Significant for Left Ears in Females: Superior Crus of Antihelix Expression (SCA) with C/P (-.267), CH with IR (-.330), SCA with No Close Friends (NCF) (.393), HR with CA (.362), and Folding of the Antihelix (FA) with CA (.374). Significant for Left Ears in Males: FA with C/P (.492).

Significant for Right Ears in Females: Darwin's Tubercle (DT) with Verbal-SAT (.362), LS with Disorganized SPQ Factor (D) (.259), CH with Odd Beliefs/Magical Thinking (OB/MT) (-.262), CH with NCF (.234), FA with OB/MT (.305), SCA with NCF (.247). Significant for Right Ears in Males: HR with CA (.259).

These data suggest that ear morphology may be a valuable complement to facial features when assessing human cognitive and personality variables. Accumulating genetic literature relating to the development of facial and ear morphologies could provide clues to the etiology of normal and abnormal behavioral states.

Disclosures: J. Cannon: None. **P.T. Orr:** None.

Poster

434. Genetic and Genomic Studies of Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 434.01/VV75

Topic: H.03. Schizophrenia

Support: T32 training grant

Title: Hippocampal subfield analysis in schizophrenia psychosis

Authors: *J. M. PEREZ, K. GLEASON, S. GHOSE, T. KIM, C. A. TAMMINGA
UT Southwestern Med. Ctr., Dallas, TX

Abstract: Schizophrenia (SZ) is one of the thirty most incapacitating conditions and affects over 67 million people worldwide; suicide occurs in 10% of those diagnosed with schizophrenia. Symptoms are persistent and often severe. They include hallucinations, delusions, thought disorder, and deficits in executive function and memory. Treatments available are not curative. 20-40% of people with SZ are entirely resistant to treatment and less than 20% completely recover. The complex symptom manifestations of SZ lack a molecular pathology. Consequently, advances in novel treatment directions are limited. We have examined postmortem human hippocampal tissue, directed by human in vivo imaging data, for molecular causes and correlates of the illness. We conceptualize psychosis as a disorder of learning and memory that critically involves dentate gyrus (DG) and CA3. We have shown considerable and consistent evidence for deficits in DG and CA3 hyperactivity with molecular and cellular changes implicating homeostatic plasticity changes within CA3. Specifically, we show increased perfusion in CA3 in schizophrenia, a correlate of neuronal activity, as well as an increase in spine density and increased protein levels of synaptic plasticity markers like GluN2B and PSD-95 in CA3 and decreased GluN1 in DG in schizophrenia. We suggest that reduced glutamatergic neurotransmission from DG may generate an increase in CA3 basal activity through homeostatic changes, possibly leading to illogical memory formation with psychotic content.
In a hypothesis generating approach, we have analyzed the transcriptome from all three hippocampal subfields from control (n=13) and schizophrenia cases (n=7 “off-drug”; n=6 “on-drug”), in a global and unbiased manner, using whole transcriptome sequencing to identify additional molecular changes, which have not been hypothesized. We have conducted differential gene expression analysis in DG, CA3, and CA1 to identify individual genes that are significantly differentially expressed between schizophrenia and control cases. We have also used the weighted gene coexpression network analysis (WGCNA) package to perform differential coexpression analysis to identify networks of genes that may differ functionally between the schizophrenia and control cases. We expect to show a network of abnormalities in the individual hippocampal subfields, which support the psychosis molecular blue print we have shown with our previous hippocampal subfield analyses.

Disclosures: J.M. Perez: None. **K. Gleason:** None. **S. Ghose:** None. **T. Kim:** None. **C.A. Tamminga:** None.

Poster

434. Genetic and Genomic Studies of Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 434.02/VV76

Topic: H.03. Schizophrenia

Support: NIMH RO1 MH098554

Title: Altered gene and protein expression of proinflammatory cytokines in the postmortem brain of schizophrenia patients

Authors: *H. ZHANG, H. RIZAVI, X. REN, G. PANDEY
Univ. of Illinois at Chicago, Chicago, IL

Abstract: Abnormalities of the immune function have been observed to be associated with the pathophysiology of schizophrenia (SZ). This is primarily based on the observation that the levels of proinflammatory cytokines, which are released from the immune cells as a result of inflammation or stress, are abnormal in the serum of patients with SZ compared with normal control (NC) subjects. However, it is not known if similar cytokines abnormalities are also present in the brain of SZ patients. In an earlier study, we reported abnormal gene expression of proinflammatory cytokines in the brain of depressed suicide victims. To further examine the involvement of inflammatory cytokines in the brain of SZ patients, we determined the protein and mRNA levels of TNF-α, IL-1β, IL-6, IL-8, IL-10, IL-13, LTA and IL-1RA in the prefrontal cortex (PFC, Brodmann area 9) of SZ patients. The study was performed in the PFC (Brodmann area 9 [BA9]) of 31 SZ patients (16 SZ suicide victims and 15 non-suicide SZ patients) and 24 NC subjects. Subject diagnosis was based on the Structured Clinical Interview for DSM-IV. The postmortem brain tissues were obtained from the Maryland Brain Collection. Protein levels of cytokines were determined by ELISA, and western blot and mRNA levels of cytokines were determined by the qPCR method. We found that the protein and mRNA expression levels of the cytokines TNF-α and IL-6 are significantly increased and those of IL-10 are significantly decreased in the PFC of SZ patients. No difference in the protein and mRNA levels of IL-1β, IL-13, and IL-1RA was observed between SZ patients and NC subjects. The protein expression levels of IL-8 were significantly decreased and those of LTA were significantly increased in SZ patients, but no significant difference in the mRNA levels of IL-8 and LTA was observed between SZ patients and NC subjects. These results suggest abnormalities of specific pro- and anti-inflammatory cytokines in the postmortem brain of SZ patients. These observations may have important implications in understanding the role of inflammatory cytokines in the pathophysiology of SZ.

Disclosures: H. Zhang: None. **H. Rizavi:** None. **X. Ren:** None. **G. Pandey:** None.

Poster

434. Genetic and Genomic Studies of Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 434.03/VV77

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIMH Grant 1R01MH111107 01A1

NIMH Grant R01 MH095995-A1

NIH Grant 5T32AG051131-02

University of Texas Medical Branch Graduate School of Biomedical Sciences Summer Internship Scholarship

Title: Effect of single nucleotide polymorphisms in fibroblast growth factor 14

Authors: *J. DI RE[1], P. A. WADSWORTH[2,3], F. LAEZZA[3,4,5]
[1]Neurosci., [2]Biochem. and Mol. Biol., [3]Pharmacol. and Toxicology, [4]Mitchell Ctr. for Neurodegenerative Dis., [5]Ctr. for Addiction Res., Univ. of Texas Med. Br., Galveston, TX

Abstract: Schizophrenia (SZ) is a complex syndrome comprising of positive, negative and cognitive symptoms with still limited medications. One barrier to medication development is the lack in knowledge of the causative links to disease heterogeneity. As the first step in understanding this complex biology, searching for functional effects of rare, naturally occurring single nucleotide polymorphisms (SNPs) in genes associated with known endophenotypes of SZ might provide new insights in disease classification and targeted therapeutic development. Recent studies have provided breakthrough results demonstrating a novel link between SZ and Fibroblast Growth Factor 14 (FGF14), a molecular component of the neuronal voltage-gated Na+ (Nav) channel complex and a regulator of synaptic plasticity. Previous studies from our group found that *Fgf14-/-* mice present with endophenotypes of SZ associated with cognitive impairment, results there were further supported by decreased expression of *FGF14* and known markers of SZ (such as *PVALB*, *GAD67* and *VGAT*) in patient-derived post-mortem tissue. We have also shown that FGF14 monomer can form homodimers that are structurally and functionally distinct from the FGF14:Nav channel complex. Building on these results, we performed a SNP analysis of FGF14 in publically available human genomic data banks and identified 83 SNPs that are predicted to change the protein coding from wild type to missense mutations or truncated products. Out of these SNPs, we are currently interrogating 9 mutations based on protein:protein interaction sites of FGF14, including sites important to its homodimer as well as the FGF14:Na_V complex. Initial screening assays of these 9 SNPs revealed that 2 SNPs, $FGF14^{F25L}$ and $FGF14^{S231N}$ exhibited decreased ability to dimerize, while other 2 SNPs $FGF14^{Y158N}$ and $FGF14^{G243D}$ exhibited opposite phenotypes ($p<.001$). In addition, all SNPs except for $FGF14^{F25L}$ have an opposite effect on complex formation between mutated FGF14 and Na_V compared to dimerization ($p<.01$). These mutations are expected to have important effects on the balance of FGF14 in its monomer and dimer form, which may in turn affect neuronal excitability and synaptic plasticity. These results may lead to a better understanding of the genetic cause of the disease, as well as new biomarker discovery and disease classification for targeted therapeutic discovery.

Disclosures: **J. Di Re:** None. **P.A. Wadsworth:** None. **F. Laezza:** None.

Poster

434. Genetic and Genomic Studies of Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 434.04/VV78

Topic: H.03. Schizophrenia

Support: NIMH IRP

Title: Resting-state connectivity in healthy adults is associated with polygenic risk for schizophrenia in dorsal frontal cortex

Authors: *M. O'BRIEN[1], M. D. GREGORY[2], M. L. ELLIOTT[2], J. P. MIKHAIEL[2], B. S. KOLACHANA[2], J. B. CZARAPATA[3], D. P. EISENBERG[4], K. F. BERMAN[5]
[1]Natl. Inst. of Mental Hlth., Bethesda, MD; [2]NIH/NIMH, Bethesda, MD; [3]NIMH/NIH, Rockville, MD; [4]Section on Integrative Neuroimaging, CBDB, [5]Section on Integrative Neuroimaging, Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: Schizophrenia is a severe psychiatric disorder characterized by delusions, hallucinations, and thought disorder, as well as negative symptoms, such as impaired cognition and blunted affect. Though schizophrenia heritability is estimated to be approximately 80%, individual single nucleotide polymorphisms (SNPs) only account for a small proportion of this heritability, suggesting the involvement of complex polygenic mechanisms. Conversely, polygenic risk scores (PRS) can be used to explain 10-20% of schizophrenia heritability. In the present study, we performed a connectome-wide association study (CWAS) of resting-state fMRI data in healthy adults to identify brain regions where schizophrenia-based polygenic risk is associated with functional connectivity. Eighty-three healthy adults (36.7+/-11.6yrs, 46 women) underwent 3T resting-state fMRI scanning and genotyping with Illumina genome-wide SNP chips. After imputation, schizophrenia PRSs were calculated for each participant using SNPs with $p<0.0001$ in the PGC-SCZ2 results using PLINK. These scores were used in a CWAS to identify regions where whole-brain connectivity was related to schizophrenia-based genetic risk. Results were thresholded at $p<0.05$, FWE-corrected. Data from significant clusters were used in post-hoc, seed-based analyses to delineate the connectivity patterns driving the association. To corroborate our results, we used data from 101 healthy young adults from the publicly available PNC dataset (19.4+/-1.1yrs, 41 women) to perform seed-based connectivity analyses of the same regions of interest. In the discovery dataset, an area of left dorsal frontal cortex (BA10) was the only region where schizophrenia polygenic risk was associated with whole-brain connectivity. A post-hoc seed-based analysis demonstrated that connectivity of BA10 to bilateral insula and inferior parietal lobules (BA39/40) drove this association. In the PNC dataset, connectivity of BA10 to the same left BA39/40 region was related to schizophrenia polygenic risk ($p<0.05$, uncorrected), though we did not replicate findings in other regions. Here, we show that

functional connectivity of BA10 with insula and BA39/40 is associated with schizophrenia polygenic risk in healthy adults. These results are consistent with prior work identifying these regions as hubs of dysfunction in schizophrenia. Our study elucidates the relationship between network functioning and continuums of polygenic risk, and suggests that these continuums exist even in healthy populations. Future work will further characterize these associations by performing similar evaluations in patients with schizophrenia.

Disclosures: M. O'Brien: None. **M.D. Gregory:** None. **M.L. Elliott:** None. **J.P. Mikhaiel:** None. **B.S. Kolachana:** None. **J.B. Czarapata:** None. **D.P. Eisenberg:** None. **K.F. Berman:** None.

Poster

434. Genetic and Genomic Studies of Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 434.05/VV79

Topic: H.03. Schizophrenia

Support: NHMRC #568807

Schizophrenia Research Institute (utilising infrastructure funding from the NSW Ministry of Health and the Macquarie Group Foundation)

Title: Polymorphism of the IL-6 SNP rs1800795 increases IL-6 production and is associated with symptom severity in chronically ill schizophrenia patients

Authors: *D. BOERRIGTER[1], T. W. WEICKERT[1,2,3], R. LENROOT[1,2], M. O'DONNELL[2], C. GALLETLY[4,5,6], D. LIU[4,5], C. SHANNON WEICKERT[1,2,3]
[1]SRL, Neura, Randwick, Australia; [2]Sch. of Psychiatry, Univ. of New South Wales, Sydney, Australia; [3]Schizophrenia Res. Inst., Randwick, Australia; [4]Discipline of Psychiatry, Sch. of Medicine, the Univ. of Adelaide, Adelaide, Australia; [5]Northern Adelaide Local Hlth. Network, Adelaide, Australia; [6]Ramsay Hlth. Care (SA) Mental Hlth., Adelaide, Australia

Abstract: Background: Recent studies have shown a significant role of cortical and peripheral cytokines in schizophrenia, specifically interleukin-6 (IL-6). Polymorphism of rs1800795 in the IL-6 gene has been associated with IL-6 production, where carriers of the mutant C allele are found to have lower IL-6 production. Blood IL-6 levels have been associated with symptom severity in (acute paranoid) schizophrenia. In this study, we tested if the rs1800795 polymorphism would be associated with blood IL-6 levels and symptom severity in a population of chronic schizophrenia patients. We hypothesized carriers of the C allele would have lower levels of IL-6 overall and may have lower symptom severity in people with schizophrenia.
Methods: Whole blood, serum and plasma from 78 controls and 95 schizophrenia or

schizoaffective patients (males and females) with mild-to-moderate symptom severity was collected, genomic DNA and messenger RNA was extracted, followed by first strand synthesis. The rs1800795 Single Nucleotide Polymorphism (SNP) was assayed and peripheral IL-6 mRNA expression levels were measured by RT-qPCR. Plasma concentrations of IL-6 were measured with the MAGPIX system. The Positive and Negative Syndrome Scale (PANSS) was administered by trained psychologists or psychometricians to obtain measures of positive, negative, total symptom severity and general psychopathology. Student t-tests were conducted for carrier vs. non-carrier analyses and ANOVA's for genotype analyses.
Results: Overall, we found that 62% of the healthy controls and 63% of the people with schizophrenia were either CG or CC (C carriers) at rs1800795 polymorphism (p=0.88). In contrast to our hypothesis, we found that C allele carriers have increased peripheral IL-6 expression (p=0.03) and increased IL-6 in plasma (p=0.04). In schizophrenia, C allele carriers had lower negative symptom scores (p=0.03) than non-carriers, no significant changes were found for positive, general or total symptom scores.
Conclusions: Our results show rs1800795 polymorphism relates to increased IL-6 production in blood in our cohort, as opposed to some previous studies. Our findings support the regulatory effect of this SNP, but we suggest that the impact of this genetic change may be context dependent. We did not find an increased incidence of the C allele carriers in schizophrenia. C allele carriers had lower negative symptom scores than non-carriers, suggesting that higher IL-6 production may work to supress negative symptoms in schizophrenia. While this finding supports our initial hypothesis of lower symptoms in C allele carriers it suggests the mechanism may be distinct than originally thought.

Disclosures: D. Boerrigter: None. **T.W. Weickert:** None. **R. Lenroot:** None. **M. O'Donnell:** None. **C. Galletly:** None. **D. Liu:** None. **C. Shannon Weickert:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Lundbeck, Astellas Pharma Inc..

Poster

434. Genetic and Genomic Studies of Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 434.06/VV80

Topic: H.03. Schizophrenia

Support: NIH Grant 5R01MH109260-02

Title: L1 retrotransposons in schizophrenia

Authors: *B. C. REINER[1], G. A. DOYLE[1], R. C. CRIST[1], A. M. PIGEON[1], R. N. LEVINSON[1], C. S. WEICKERT[2], G. TURECKI[3], T. N. FERRARO[4], W. H. BERRETTINI[1]
[1]Ctr. for Neurobio. and Behavior, Univ. of Pennsylvania, Philadelphia, PA; [2]Neurosci. Res.

Australia, Sydney, Australia; [3]Psychiatry, McGill, Montreal, QC, Canada; [4]Biomed. Sci., Cooper Med. Sch. of Rowan Univ., Camden, NJ

Abstract: Schizophrenia (SZ) is a neurodevelopmental, partially inherited, chronic psychotic disorder affecting 0.5 - 1.0% of the population. While current antipsychotic medications can treat the positive symptoms of SZ, including hallucinations and delusions, there is no FDA-approved therapy for the negative symptoms of SZ, including apathy, avolition and poverty of thought. Despite evidence that the heritability of SZ is ~64%, the identification of common alleles, via leukocyte DNA, with even a moderate increase in the risk for SZ has proven difficult. Therefore, alternative approaches to identifying genetic variants associated with SZ are needed. We examined the role of neuronal somatic mutation in increasing the risk for developing SZ. While somatic DNA variation wouldn't explain the heritability of SZ, it would explain the "environmental" risk of SZ. We focused on the study of somatic neuronal DNA variation mediated by long interspersed nuclear element 1 (LINE1 or L1) retrotransposons, a type of mobile DNA element, with the potential of disrupting genes, which may increase the risk for SZ. DNA was isolated from NeuN+ dorsolateral prefrontal cortex neuronal nuclei from 48 SZ persons and 48 age, sex and ethnicity matched controls. L1 specific DNA was amplified using PCR, and L1 amplicons were subject to next generation sequencing. Sequence data will then be aligned to the human reference genome to identify neuronally expressed genes with de novo L1 insertions, and bioinformatics will be used to determine the relevant biological pathways.

Disclosures: B.C. Reiner: None. **G.A. Doyle:** None. **R.C. Crist:** None. **A.M. Pigeon:** None. **R.N. Levinson:** None. **C.S. Weickert:** None. **G. Turecki:** None. **T.N. Ferraro:** None. **W.H. Berrettini:** None.

Poster

434. Genetic and Genomic Studies of Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 434.07/VV81

Topic: H.03. Schizophrenia

Support: NIH Intramural Research Program

Title: Dysbindin regulates mitochondrial fission in hippocampal excitatory neurons

Authors: *J. ZHAO[1], Z. LI[2]
[1]NIMH/NIH, Bethesda, MD; [2]NIMH, Bethesda, MD

Abstract: Background: Mitochondria have several functions, such as the production of energy and reactive oxygen species (ROS) and calcium buffering, which are essential for neuronal activity and connectivity. Mitochondria form networks in the cell. The mitochondrial network is

dynamically regulated by mitochondrial fission and fusion, which are required for the biogenesis and quality control of mitochondria. Mitochondria dynamics is controlled by mitochondrial fission and fusion proteins, such as MFN-1, DLP1, FIS-1 and so on. Earlier reports have implicated a possible linkage between mitochondrial dysfunction and schizophrenia. However, how schizophrenia risk genes regulate mitochondrial functions and mitochondrial network have not been investigated. Result: In our study, we investigated the role of the human schizophrenia susceptibility gene DTNBP1, encoding dysbindin protein, in mitochondria dynamics. We found that a lack of dysbindin in excitatory neurons results in longer mitochondria and less mitochondrialin postsynapses. These impariments were rescued by dysbindin-1c.Dysbindin significantly changed the subcellular oligomeration of DLP1 and acvitity-induced mitochondrial fission. Conclusion: Our results indicate that dysbindin regulates mitochondrial dynamics and function through DLP1, and therefore dysbindin deficiency may contribute to impaired neural network activity in schizophrenia by causing mitochondrial defects.

Disclosures: J. Zhao: None. **Z. Li:** None.

Poster

434. Genetic and Genomic Studies of Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 434.08/VV82

Topic: H.03. Schizophrenia

Support: NIH Grant R01MH094358

Title: Chromatin immunoprecipitation followed by deep sequencing reveals differential repressive chromatin sites in schizophrenia post mortem brain

Authors: B. M. FEINER[1], J. K. MELBOURNE[2], C. ROSEN[3], *R. P. SHARMA[4]
[1]Psychiatry, UIC Psychiatry, Chicago, IL; [2]Psychiatry, [3]Univ. of Illinois At Chicago, Chicago, IL; [4]Dept Psychiatry, Univ. of Illinois at Chicago, Chicago, IL

Abstract: Aberrant chromatin modification levels have repeatedly been shown in the brains and immune cells of patients with schizophrenia. However, the majority of these studies measured total levels of histone modifications from populations of cells, which only allows for quantification of a total increase or decrease without revealing the genomic locations of the modified chromatin. Chromatin immunoprecipitation followed by deep sequencing (ChIP-Seq) allows us to generate a map of the genome that includes differential sites of a chromatin modification of interest. By targeting di-methylated lysine 9 of histone 3 (H3K9me2), a modification that has never been done before in this way in human post-mortem brain, we have discovered over one hundred highly significant differential sites in schizophrenia prefrontal cortex (N=15) when compared to controls (N=15). Bioinformatics and pathway analysis revealed

significant enrichments of genes previously determined to be involved in biological areas important to schizophrenia, such as the synapse or formation of the cortex. To determine functional significance, genes with promoters that had significantly increased or decreased H3K9me2 levels were selected and qPCR was performed. mRNA expression analysis revealed that expression levels correlated well with chromatin modification levels, i.e. genes with H3K9me2-enriched promoters had less mRNA expression and vice versa. Utilization of this method has already provided a number of candidate genes for potential future studies (e.g. CRISPR-directed promoter methylation in cells), and it could be a useful tool in understanding a complex and heterogeneous disorder such as schizophrenia.

Disclosures: B.M. Feiner: None. **J.K. Melbourne:** None. **C. Rosen:** None. **R.P. Sharma:** None.

Poster

434. Genetic and Genomic Studies of Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 434.09/VV83

Topic: H.03. Schizophrenia

Support: NIH R01 MH097803

The Danish Council for Independent Research

The Lundbeck Foundation

Title: Sleep deprivation induces expression of serotonin 2A receptors in the frontal cortex of mice, in an Egr3-dependent manner

Authors: *A. VANNAN[1,2], K. T. MEYERS[1,2], A. M. MAPLE[2], D. I. ELIZALDE[2], X. ZHAO[1,2], A. OVERGAARD[3,4], G. M. KNUDSEN[3,4], A. L. GALLITANO[2,1]
[1]Interdisciplinary Grad. Program in Neurosci., Arizona State Univ., Tempe, AZ; [2]Basic Med. Sci., Univ. of Arizona Col. of Med., Phoenix, AZ; [3]Neurobio. Res. Unit, Copenhagen Univ. Hosp. Rigshospitalet, Copenhagen, Denmark; [4]Fac. of Hlth. and Med. Sci., Univ. of Copenhagen, Copenhagen, Denmark

Abstract: Genetic and environmental factors interact to influence an individual's risk for schizophrenia. Immediate early gene transcription factors (IEG-TFs) are rapidly activated by environmental events such as stress and, in turn, dictate the molecular response of the brain to these events. Dysfunction of IEG-TFs may provide a mechanism to explain the dual genetic and environmental contributions to risk for mental illness. Indeed, the IEG-TF *early growth response 3* (*Egr3*) has been associated with schizophrenia in multiple populations. *Egr3* expression is decreased in the postmortem brains of patients with schizophrenia. We have previously

demonstrated a link between *Egr3* and the schizophrenia candidate gene, *Htr2a*, which encodes the serotonin 2A receptor (5-HT_{2A}R). *Egr3-/-* mice display behavioral abnormalities characteristic of a schizophrenia-like phenotype and have reduced cortical 5-HT_{2A}R levels. Likewise, patients with schizophrenia also display a decrease in cortical 5-HT_{2A}Rs. Our prior work has shown that sleep deprivation (SD), a mild stressor known to activate *Egr3*, increases *Htr2a* expression in the cortex of WT, but not *Egr3-/-*, mice, detected by quantitative real-time polymerase chain reaction (qRT-PCR). In the current study, we examined the cortical regions in which SD activates *Htr2a* expression, and whether this induction requires *Egr3*, using *in situ* hybridization (ISH). In addition, we examined whether this increased expression in response to SD resulted in an increase in level of cortical 5-HT_{2A}R protein detected by 3H-MDL100907 binding. Our results from both ISH and radioligand binding indicate that SD increases 5-HT_{2A}R expression in regions of the frontal cortex of WT mice, but this induction is absent in mice lacking *Egr3*. These findings provide insight into stress-induced cortical abnormalities that are also observed in patients with schizophrenia.

Disclosures: **A. Vannan:** None. **K.T. Meyers:** None. **A.M. Maple:** None. **D.I. Elizalde:** None. **X. Zhao:** None. **A. Overgaard:** None. **G.M. Knudsen:** None. **A.L. Gallitano:** None.

Poster

434. Genetic and Genomic Studies of Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 434.10/VV84

Topic: H.03. Schizophrenia

Title: Exploring polygenic risk in cognitive trajectory subgroups of schizophrenia: Unexpected differences in correlations with cognition

Authors: *S. R. ZAIDMAN[1], E. GIANGRANDE[2], D. R. WEINBERGER[3], K. F. BERMAN[4], D. DICKINSON[5]
[1]Clin. and Translational Neurosci. Br., NIMH, Bethesda, MD; [2]Psychology, Univ. of Virginia, Charlottesville, VA; [3]Lieber Inst. For Brain Develop., Baltimore, MD; [4]Section on Integrative Neuroimaging, Natl. Inst. of Mental Hlth., Bethesda, MD; [5]Clin. and Translational Neurosci. Br., Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: Background:
The purpose of this study was to evaluate associations between cognition and risk profile scores (RPS) for schizophrenia patients divided into three subgroups determined by premorbid and fluid intelligence.
Methods:
550 people with schizophrenia, 432 of their unaffected siblings, and 1127 community controls provided demographic and clinical information, completed cognitive and symptom assessments

and provided blood for genetics analyses, as part of the NIMH Study of Schizophrenia Genetics. We performed cluster analyses (SPSS) in the schizophrenia cases to identify cognitive subgroups, using only “premorbid” (WRAT) and “current” (WAIS) IQ as clustering indicators, with the hypothesis that different patterns of performance identify subgroups with distinct trajectories of cognitive development. Schizophrenia RPS for all individuals were calculated at 10 thresholds based on illness-associated genetic variants identified by the multi-national Psychiatric Genetics Consortium. Across RPS thresholds, we used planned hierarchical regression to test the association of RPS with general cognitive ability (“g”) in the derived cognitive subgroups, controlling for age, sex, and population stratification.

Results:

Based on 1000 runs, three cluster solutions were the most frequent result, suggesting one subgroup with high scores on both WRAT and IQ (Cognitively Stable), one with low scores on both WRAT and IQ (Pre-Adolescent Impairment), and one with high scores on the WRAT and low IQ (Adolescent Decline). The Cognitively Stable subgroup showed significant associations between RPS and “g” at six of 10 RPS thresholds (e.g., at RPS_0.05 p=.002; R^2=0.047). The Pre-Adolescent Impairment subgroup showed a pattern of similar but non-significant associations. However, the Adolescent Decline subgroup showed no evidence of association of schizophrenia RPS with “g” at any threshold.

Conclusions:

Cognitive impairment is recognized as a core feature of schizophrenia. These analyses suggest that for some individuals with schizophrenia, common genetic risk for the condition is also predictive of cognitive impairment. However, for another subgroup, polygenic risk for schizophrenia is entirely “decoupled” from cognition. Differences in cognitive profile and in the association of cognitive performance with common genetic risk for schizophrenia may point to important distinctions in etiology. Ongoing analyses aim to further characterize differences between these subgroups, particularly in terms of environmental factors that could impact development and cognition.

Disclosures: S.R. Zaidman: None. **E. Giangrande:** None. **D.R. Weinberger:** None. **K.F. Berman:** None. **D. Dickinson:** None.

Poster

434. Genetic and Genomic Studies of Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 434.11/VV85

Topic: H.03. Schizophrenia

Support: AZV 15-29370A

Biocev CZ.1.05/1.1.00/02.0109

Grant RVO: 67985823

NPUII Grant LQ1604

Title: Genetic analysis of regulatory elements of the NMDARs in schizophrenia

Authors: *A. BALIK[1], J. CERNY[2], T. RAUSCH[3], V. BENES[3], S. HRYCHOVA[4], J. HORACEK[6], L. VYKLICKY[5]
[1]Inst. of Physiology, ASCR, Vestec, Czech Republic; [2]Inst. of Biotechnology, CAS, Vestec, Czech Republic; [3]EMBL, Heidelberg, Germany; [4]BIOCEV, Inst. of Physiology, CAS, Vestec, Czech Republic; [5]Inst. of Physiology, CAS, Prague, Czech Republic; [6]Natl. Inst. of Mental Hlth., Klecany, Czech Republic

Abstract: Schizophrenia (SCH) is a serious neuropsychiatric disorder with a severe impact on patients' lives. Based on the experimental models it was concluded that altered (decreased) activity of N-methyl-D-aspartate receptor (NMDAR) induces behavior resembling the clinical manifestations of schizophrenia. Therefore, we aimed to determine gene variants of the glutamate receptor regulatory elements and their contribution to the emergence and development of schizophrenia. We were focused on an in-depth analysis of genes encoding NMDARs in selected subgroup of SCH patients showing strong and unique disease phenotype linked to early onset of psychosis. Since promoter regions of gene is the key element that regulates the production of new receptors and thus may be directly responsible for lower gene expression we analyzed distribution and frequency of SNPs in the extended promoter regions of NMDARs. Using NimbleGene SeqCap EZ system we have observed and subsequently analyzed more than 170 SNPs in promoter regions of six NMDAR genes. We found two times higher frequency of SNPs in promoter regions of GluN2B and 3B subunits compared to the other ionotropic glutamate receptors we analyzed. This indicates a greater probability of a change in the activity of these promoters which could also cause a decrease in the overall expression of these subunits. We also found several SNPs in GluN3A promoter region with different distribution between our group of SCH patients and control healthy volunteers. Moreover, one fourth of SCH patient samples carried specific combinations of SNPs in promoter region that we did not observed in control samples. In summary, our data confirmed previous observations that the genetic information of regulatory elements of NMDARs is impacted in SCH. We assume that our data could contribute to the identification of the set of genetic markers indicating higher susceptibility to emergence and development of schizophrenia.

Disclosures: A. Balik: None. **J. Cerny:** None. **T. Rausch:** None. **V. Benes:** None. **S. Hrychova:** None. **J. Horacek:** None. **L. Vyklicky:** None.

Poster

434. Genetic and Genomic Studies of Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 434.12/VV86

Topic: H.03. Schizophrenia

Title: Using random forest machine learning to identify multi-genetic influences on neural activity underlying motivation

Authors: *Q. CHEN[1], R. W. LEFCO[1], R. E. STRAUB[1], K. K. NICODEMUS[2], D. R. WEINBERGER[1], C. F. ZINK[1]
[1]Lieber Inst. For Brain Develop., Baltimore, MD; [2]Univ. of Edinburgh, Edinburgh, United Kingdom

Abstract: Determining genetic influences on human neural activity patterns is likely highly relevant to clinical advances in the treatment of heritable mental illnesses, such as schizophrenia. Traditional techniques in "imaging genetics" - the correlation of genetic variation with neuroimaging signals to speak to cellular/molecular influences - are limited by testing only the effects of one or a few variants at a time without accounting for multi-genetic effects/epistasis, and the variations selected may have limited involvement. We overcome these challenges using an innovative imaging genetics approach that utilizes Random Forest (RF) machine learning to perform a multiple regression. RF is a high-dimensional, non-parametric predictive model composed of a collection of regression (or classification) trees. RF has the potential to uncover interactions among multiple genes that may exhibit only weak main effects, thus providing the percent of variance explained in a signal by multi-genetic effects. It has the advantage of being able to handle large numbers of genetic variants. It also provides an importance score for each variable, rendering RF capable of identifying genetic variations that are most predictive of a given neural activity, which can narrow the focus of future investigations on particular promising genes. Here, we focus on motivation neural activity because lack of motivation is a debilitating symptom of schizophrenia that is generally unimproved by current anti-psychotic medications. Using fMRI, we precisely isolate motivation-related striatal activity in 86 healthy Caucasian adults. Genetic variations of interest were limited to known, common (MAF > 0.15), functional (significant eQTLs) single nucleotide polymorphisms (SNPs) in dopaminergic, glutamatergic, GABAergic, and cholinergic signaling genes because of their well-characterized roles in the striatum. Separate RF regressions were conducted on "gene-sets" categorized by neurotransmitter system. We report the gene-sets that predict a significant amount of variance in the imaging signal and the genes/genetic variations ranked as most influential. We find a significant multi-genetic effect of SNPs in GABA signaling genes on motivation-related striatal activity, with SNPs in GAD2 and subunits of the GABA-A receptor being most important. Our findings implicate the GABA signaling pathway as particularly important for motivation-evoked activity

in the striatum and provide evidence for the utility of the usage of RF multiple regression in imaging genetics to determine multi-genetic/epistatic influences on human neural activity.

Disclosures: Q. Chen: None. **R.W. Lefco:** None. **R.E. Straub:** None. **K.K. Nicodemus:** None. **D.R. Weinberger:** None. **C.F. Zink:** None.

Poster

434. Genetic and Genomic Studies of Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 434.13/VV87

Topic: H.03. Schizophrenia

Title: Maternal immune activation impacts schizophrenia-related microRNA related to neuroinflammation

Authors: *S. K. AMOAH[1], B. A. RODRIGUEZ[1], C. N. LOGOTHETIS[1], T. R. YELLOWHAIR[2], J. P. WEICK[1], L. L. JANTZIE[1,2], N. MELLIIOS[1]
[1]Dept. of Neurosciences, [2]Dept. of Pediatrics, Univ. of New Mexico HSC, Albuquerque, NM

Abstract: Schizophrenia and bipolar disorder are heterogeneous psychiatric conditions with elusive pathogenesis. Given that protein-coding genes inhabit less than half of the human brain transcriptome it has been speculated that actively transcribed non-coding RNAs (ncRNAs) could contribute significantly to the pathophysiology of psychiatric disease. microRNAs (miRNAs) are a subtype of small evolutionarily conserved ncRNAs that have been shown to regulate several aspects of brain development and synaptic plasticity. Maternal immune activation (MIA) during pregnancy can increase the prevalence of neurodevelopmental disorders, such as schizophrenia, in offspring. We hypothesized that inflammation-related miRNAs (inflammiRs) are altered in the brains of psychiatric patients, and that this could be related to prenatal inflammation. We also tested that MIA in rats would yield similar changes. Employing methods that selectively detect the mature miRNA transcript, we screened and validated numerous differentially expressed inflammiRs altered in postmortem brains from the orbitofrontal cortex (OFC) of schizophrenia and bipolar disorder patients. Results indicate that miR-223 was the most robustly increased inflammiR transcript in schizophrenia brains ($p < .001$). Subsequently, we observed reductions in several of its validated neuronal targets, such as N-methyl D-aspartate receptor subtype 2B (NR2B) and Glutamate receptor 2 (GRIA2). Interestingly, levels of miR-223 were positively correlated with the expression of inflammatory genes, such as Alpha 1-antichymotrypsin (AACT or Serpina3) ($p < .01$) and interleukin 6 (IL-6), and yet showed significant negative associations with its validated targets NR2B ($p < .05$) and GRIA2 expression ($p < .01$). Notably, miR-223 levels were upregulated in embryonic day 17 (E17) brains following intraperitoneal (i.p.) administration of lipopolysaccharide (LPS, 100μg/kg, once daily) on E15 and E16, corroborating our clinical data ($p < .001$). Together, our data show that altered expression of inflammiRs in

schizophrenia can influence NMDA and glutamate receptor levels in human orbitofrontal cortex and could be associated with maternal immune activation. Experiments using human stem cell-derived neuronal cultures and additional rodent models of MIA are underway to evaluate effects of knockdown of miR-223 on microglia and synaptic plasticity.

Disclosures: **S.K. Amoah:** None. **B.A. Rodriguez:** None. **C.N. Logothetis:** None. **T.R. Yellowhair:** None. **J.P. Weick:** None. **L.L. Jantzie:** None. **N. Melliios:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.01/VV88

Topic: H.03. Schizophrenia

Support: NIMH Grant MH105660

Title: Study of the functional importance of DISC1 Serine 713/710 phosphorylation in brain development and cognitive behavior in human subjects and in novel mouse models

Authors: *H. JAARO-PELED[1], N. GAMO[1], Y. HORIUCHI[1], S. ISHII[4], K. ISHII[1], K. NISHIHARA[4], Z. HO[2], S. KULASON[3], S. MORI[2], D. SCHRETLEN[1], H. OKANO[4], T. RATNANATHER[3], M. MILLER[3], K. ISHIZUKA[1], A. SAWA[1]
[1]Psychiatry and Behavioral Sci., [2]Radiology, [3]Biomed. Engin., Johns Hopkins Univ., Baltimore, MD; [4]Keio Univ., Tokyo, Japan

Abstract: DISC1 is involved in many aspects of brain development and function, however, the regulation of its function is poorly understood. We previously identified phosphorylation of mouse Disc1 Serine 710 (S710) as a critical switch from neural progenitor proliferation to migration in the developing cortex using *in utero* electroporation. This study's objectives are (1) to test the significance of this finding in humans using a multifaceted approach and (2) to use newly generated mouse models in which Disc1 S710 has been mutated to test the causality of this phosphorylation in brain development and behavior.
For the human part of the study we collected olfactory cells and fibroblasts from psychotic subjects and from healthy controls, and performed neurocognitive assessments and structural MRI. Using olfactory neurons we found that psychotic subjects had lower phosphorylation levels of DISC1 at S713 (homologous to mouse S710) than controls. Using induced pluripotent stem cells derived from the subjects whose olfactory neurons showed low phosphorylation of DISC1 at S713, we detected delayed neuronal maturation. We also demonstrated a positive correlation between the phosphorylation level in olfactory neurons and the thickness of the anterior cingulate cortex and working memory/attention.
For the mouse part of the study we generated point mutants at S710 using CRISPR technology.

We generated a phospho dead mutant, S710Alanine and a phospho-mimetic mutant, S710Glutamate. The mice are viable and grossly normal even as homozygotes. We performed *ex-vivo* MRI to test the effect of S710A on brain anatomy at P28 (juvenile) and P70 (young adulthood). We found several abnormalities including smaller frontal association cortex, which we are following on with immunohistochemistry. We are also performing neuronal proliferation and migration assays and behavioral testing focusing on cognitive function.
In conclusion, phosphorylation of DISC1 at Serine 713/710 is critical for maturation of neural progenitors, and correlates with human brain development and behavior relevant to psychosis and cognitive deficits. Using our novel mouse models we aim to show causality of this phosphorylation towards these phenotypes.

Disclosures: H. Jaaro-Peled: None. **N. Gamo:** None. **Y. Horiuchi:** None. **S. Ishii:** None. **K. Ishii:** None. **K. Nishihara:** None. **Z. Ho:** None. **S. Kulason:** None. **S. Mori:** None. **D. Schretlen:** None. **H. Okano:** None. **T. Ratnanather:** None. **M. Miller:** None. **K. Ishizuka:** None. **A. Sawa:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.02/VV89

Topic: H.03. Schizophrenia

Support: NIH Grant MH057440

NIH Grant MH109199

Title: Medial septum differentially regulates spontaneous dopamine neuron activity in the ventral tegmental area and substantia nigra pars compacta via distinct neurochemical pathways

Authors: *D. M. BORTZ, A. A. GRACE
Dept. of Neuroscience, Psychiatry, and Psychology, Ctr. for Neurosci., Univ. of Pittsburgh, pittsburgh, PA

Abstract: Disruptions in dopamine (DA) signaling are central to the pathophysiology of several major psychiatric disorders. Thus, the discovery of novel therapeutic approaches that normalize DA signaling is a major focus of research. One pathway that is critical for DA regulation originates from the ventral subiculum (Vsub) of the hippocampus and controls ventral tegmental area (VTA) DA neuron activity. A relatively unstudied regulator of hippocampal function is the medial septum (MS); a sub-region of the cholinergic basal forebrain that innervates the hippocampus, including the Vsub, via cholinergic and GABAergic projections, drives hippocampal theta rhythms, and affects goal-directed behavior. Despite this, it has never been

determined if the MS is an afferent regulator of the midbrain DA system, and therefore may be a novel therapeutic treatment target. This question was addressed by infusing NMDA (0.75 µg/0.2 µL) into the medial septum of anesthetized male Sprague-Dawley rats and recording dopamine neuron activity in the VTA and substantia nigra pars compacta (SNc), an adjacent DA region also implicated in psychopathology. MS activation produced a prolonged 71% increase in the number of spontaneously active DA cells in the VTA, and an opposing 40% decrease in the number of active DA cells in the SNc, compared to vehicle infusions. Effects in both the VTA and SNc were dependent on the Vsub as infusion of TTX (1 µM /0.5 µL) into the Vsub reversed population activity changes in both regions. Interestingly, effects in the VTA and SNc were precipitated by different neurotransmitter projections from the MS, with infusion of muscarinic antagonist scopolamine (8 µg/1.0 µL) into the Vsub preventing MS activation-induced DA population activity changes in the VTA, but not the SNc; whereas Vsub infusions of GABAergic antagonist bicuculline (12.5 ng/0.5 µL) prevented population activity changes in the SNc, but not the VTA. Furthermore, the ventral pallidum (VP) was necessary for effects in both the VTA and SNc; however, bicuculline (1 ng/0.2 µL) infusion into the rostral VP selectively prevented population activity increases in the VTA, while bicuculline infusion into the caudal VP selectively prevented population activity decreases in the SNc. These findings demonstrate that distinct septohippocampal pathways play a key modulatory role in the regulation of meso-striatal DA transmission originating from both the VTA and SNc, indicating this pathway may present novel therapeutic approaches for several psychological disorders.

Disclosures: D.M. Bortz: None. **A.A. Grace:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.03/VV90

Topic: H.03. Schizophrenia

Support: Pritzker Neuropsychiatric Research Consortium

NIH: R01MH104261

ONR: N00014-15-1-2224

Hope for Depression Research Foundation

Title: Molecular mapping of the human thalamus. Relationship with thalamocortical wiring and role in schizophrenia

Authors: *R. CALZAVARA[1], H. AKIL[1], J. D. BARCHAS[2], W. E. BUNNEY[3], F. S. LEE[2], R. M. MYERS[4], A. F. SCHATZBERG[5], S. J. WATSON[1]

[1]Mol. and Behavioral Neurosci. Inst., Mol Beh Neurosci In Univ. of Michigan Med. Sch., Ann Arbor, MI; [2]Dept. of Psychiatry, Dept. of Psychiatry Weill Cornell Med. Col., New York, NY; [3]Psychiatry and Human Behavior, Psychiatry and Human Behavior Univ. of California - Irvine, Orange, CA; [4]Hudson Alpha Inst. for Biotech., Hudson Alpha Inst. for Biotech., Huntsville, AL; [5]Dept. of Psychiatry and Behavioral Sci., Dept. of Psychiatry and Behavioral Sciences, Stanford Univ., Stanford, CA

Abstract: The thalamus is a key structure for relaying and integrating multi-functional information for perception, action and cognition within the cortex. Anatomical and functional data indicate that thalamo-cortical connectivity is impaired in schizophrenia. In particular, dysfunctional connectivity with frontal lobe areas, such as the dorsolateral prefrontal cortex (DLPFC) and the anterior cingulate cortex (ACC), may represent a potential neural substrate for cognitive dysfunction. As a first step toward a cell type specific model of cortico-thalamic fine wiring in schizophrenia, we investigated the basic neurochemical and molecular mapping of the thalamus in control subjects. In particular, we focused on the nuclei connected to frontal cortex, such as the medial dorsal (MD) nucleus. We identified molecular markers of thalamic neurons and we used in situ hybridization to investigate the relative cell expression and distribution of these markers in the thalamus. Preliminary results indicate a complex neurochemical map in which some markers have complementary or similar distributions. In particular, some marker distributions seem to overlap in specific thalamic nuclei, such as observed for the cell clusters in the MD nucleus. This may indicate that thalamic cells can be identified by a unique set of molecular markers. The relative expression of these markers and their distributions in the thalamic relay nuclei is thought to be associated with cell development and neurotransmitter function, and therefore can be related to specific thalamo-cortical circuits. The major goal of our study is to test whether these thalamo-cortical circuits, and potential selective cell types, are particularly affected in schizophrenia. Thus, this gene-expression map of the human thalamus would guide our next steps in the investigation of thalamic gene-expression patterns in schizophrenia.

Disclosures: **R. Calzavara:** Other; The author is member of the Pritzker Neuropsychiatric Disorders Research Consortium, which is supported by the Pritzker Neuropsychiatric Disorders Research Fund L.L.C. **H. Akil:** Other; The author is member of the Pritzker Neuropsychiatric Disorders Research Consortium, which is supported by the Pritzker Neuropsychiatric Disorders Research Fund L.L.C. **J.D. Barchas:** Other; The author is member of the Pritzker Neuropsychiatric Disorders Research Consortium, which is supported by the Pritzker Neuropsychiatric Disorders Research Fund L.L.C. **W.E. Bunney:** Other; The author is member of the Pritzker Neuropsychiatric Disorders Research Consortium, which is supported by the Pritzker Neuropsychiatric Disorders Research Fund L.L.C. **F.S. Lee:** Other; The author is member of the Pritzker Neuropsychiatric Disorders Research Consortium, which is supported by the Pritzker Neuropsychiatric Disorders Research Fund L.L.C. **R.M. Myers:** Other; The author is member of the Pritzker Neuropsychiatric Disorders Research Consortium, which is supported by the Pritzker Neuropsychiatric Disorders Research Fund L.L.C. **A.F. Schatzberg:** Other; The author is member of the Pritzker Neuropsychiatric Disorders Research Consortium, which is supported by the Pritzker Neuropsychiatric Disorders Research Fund L.L.C. **S.J. Watson:** Other;

The author is member of the Pritzker Neuropsychiatric Disorders Research Consortium, which is supported by the Pritzker Neuropsychiatric Disorders Research Fund L.L.C..

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.04/VV91

Topic: H.03. Schizophrenia

Support: NIH R01 MH100066

Title: Immune activation across the visuospatial working memory circuit in schizophrenia

Authors: *A. R. LOCKER[1,2], E. SIPPLE[2], P. LIDSTONE[2], D. VOLK[2]
[1]Psychiatry, UPMC, Pittsburgh, PA; [2]Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Cognitive impairments, including deficits in visuospatial working memory, are debilitating features of schizophrenia. Immune activation, indicated by higher transcript levels for cytokines and interferon-induced transmembrane proteins (IFITMs), have been reported in the prefrontal cortex (area 9) in schizophrenia. The transcription factor nuclear factor-κB (NF-κB) regulates the expression of these IFITM variants as well as the same cytokines that are overexpressed in schizophrenia. We recently found higher mRNA levels for multiple NF-κB family members (NF-κB1, NF-κB2, RelA, cRel) and initiation receptors (IL-1R, TNFR) in the canonical NF-κB activation pathway in area 9 of schizophrenia subjects. Consequently, we examined whether levels of these NF-κB-related markers are higher throughout cortical regions in the visuospatial working memory circuit, including dorsolateral prefrontal (area 46), posterior parietal (area 7), and visual (areas 18 and 17) cortices, in 20 schizophrenia subjects relative to 20 unaffected comparison subjects using quantitative PCR. Overall, schizophrenia subjects had significantly higher mRNA levels for all measured NF-κB-related markers relative to unaffected comparison subjects. Statistically significant regional differences were also seen for many measured NF-κB family members. Region × diagnosis interactions were significant (or nearly significant) for all NF-κB family members. Post-hoc tests revealed that in schizophrenia subjects, most NF-κB-related markers were significantly (or nearly significantly) elevated in each cortical region relative to unaffected comparison subjects. However, post-hoc tests also revealed that in most cases, schizophrenia subjects, but not unaffected comparison subjects, displayed higher levels of NF-κB-related markers in primary and secondary visual cortex relative to posterior parietal and prefrontal cortex. These findings suggest that 1) levels of NF-κB-related markers are elevated across the entire visuospatial working memory circuit in schizophrenia subjects, and 2) these increases may be most prominent in visual cortex. The presence of immune activation across cortical regions may disrupt cortical circuitry function in schizophrenia. Determinations of

disturbances in the precise pathways that underlie cognitive dysfunction in schizophrenia may ultimately aid in the discovery of therapeutic treatments of this disorder.

Disclosures: **A.R. Locker:** None. **E. Sipple:** None. **P. Lidstone:** None. **D. Volk:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.05/VV92

Topic: H.03. Schizophrenia

Support: Stanley Foundation

NIMH R01 MH074983

NIMH R21 MH091774

Brain and Behavior Research Foundation Young Investigator Grant

Dr. Victor Milstein Clinical and Research Fund for Psychiatry IUSM

Title: Relationship of auditory event related potentials with magnetic resonance spectroscopy metabolites in early stage psychosis

Authors: L. BARTOLOMEO[1], A. M. WRIGHT[2], R. MA[2], T. A. HUMMER[3], M. M. FRANCIS[3], A. C. VISCO[3], N. MEHDIYOUN[3], A. R. BOLBECKER[1], W. P. HETRICK[1], U. DYDAK[2], *B. F. O'DONNELL[4], A. BREIER[3]
[1]Dept Psychological and Brain Sci., Indiana Univ., Bloomington, IN; [2]Sch. of Hlth. Sci., Purdue Univ., Lafayette, IN; [3]Dept. of Psychiatry, Indiana Univ. Sch. of Med., Indianapolis, IN; [4]Dept Psychol, Indian Univ., Bloomington, IN

Abstract: Event-related potential (ERP) deficits in the mismatch negativity (MMN) elicited by deviant tones in a series and the auditory steady state response (ASSR) to 40 Hz stimulation are usually found in schizophrenia (SZ), but the neural basis of these disturbances are not well characterized. *In vivo* magnetic resonance spectroscopy (MRS) have shown alterations in several metabolites, including increased glutamatergic levels (Merritt et al, 2016) and reduced N-acetylaspartate (NAA) levels in SZ (Brugger et al, 2010). The relationship of electrophysiological responses to brain metabolites could provide insights into the neurochemical basis of scalp recorded ERP abnormalities early in the course of psychosis. In the present study, the correlation of MMN and the 40 Hz ASSR to glutamine, N-acetylaspartate, and other metabolites measured with magnetic resonance spectroscopy (MRS) were examined in patients with early stage psychosis. Thirty one individuals within 3 years of the onset of a non-affective

psychotic disorder were tested. MMN amplitude was measured from ERPs to duration deviant tones and ASSR power to 40 Hz click trains (500 ms duration) at midline electrode sites. The MRS volume of interest (VOI) was placed medially in the frontal cortex (size = 2x2x2 cm^3; TE = 30ms; TA = 4min) using a Siemens 3T TIM Trio whole-body scanner with a 32-channel head coil. N-acetylaspartate (NAA), glutamine (Glx), creatine (Cre), myo-inositol (MI) and choline (Cho) were measured. Pearson correlation coefficients were computed between ERP measures and metabolite levels with two-tailed *p* values for significance testing. MMN was correlated with Cre (r = .44, p = .01), Glx (r = .40, p = .03), NAA (r = .49, p < .01) and Cho (r = .44, p = .01) indicating that higher metabolite levels were associated with smaller (less negative) MMN responses. ASSR showed trends (r < .10) for correlations with Glx (r = .30), suggesting that higher levels of Glx was associated with greater 40 Hz ASSR power. In these early stage patients with psychosis, higher metabolic signals were strongly related to reduced MMN amplitude. Notably, elevations of Glu have been reported in schizophrenia (Merritt et al, 2016) and increased concentrations of Cre in the frontal lobe of children with schizophrenia (O'Neill et al, 2004).

Disclosures: L. Bartolomeo: None. **A.M. Wright:** None. **R. Ma:** None. **T.A. Hummer:** None. **M.M. Francis:** None. **A.C. Visco:** None. **N. Mehdiyoun:** None. **A.R. Bolbecker:** None. **W.P. Hetrick:** None. **U. Dydak:** None. **B.F. O'Donnell:** None. **A. Breier:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.06/WW1

Topic: H.03. Schizophrenia

Support: 15K19735

Title: Facial emotion processing in schizophrenia using newly created facial stimuli by japanese actors; fMRI study

Authors: *N. ORIBE[1,2], H. KUGA[2], H. TAKAO[2], S. FUKUSHIMA[2], Y. HIRANO[1], T. ONITSUKA[1], T. UENO[2]
[1]Kyushu Univ., Fukuoka, Japan; [2]Hizen Psychiatric Ctr., Yoshinogari, Japan

Abstract: Background: Patients with schizophrenia have been associated with deficits in many aspects of social cognition. Especially, perception of face and/or facial expression is impaired through the course of the illness and that might result in the severe social dysfunction. Functional MRI has been used to detect the key region of this deficit. However, the results from previous studies have not been always consistent partly because of the differences in cultural background. It is commonly more difficult to distinguish the facial emotion in unfamiliar ethnic people then

in the familiar. In the current study, we adopted the facial emotion stimuli, which are played by Japanese actor/actress and evaluated as showing the intended emotions, during the measurement of fMRI.
Methods: Six schizophrenia patients and 7 healthy controls participated in two tasks; Discrimination Task and Emotion Task In the Discrimination task, forty pairs of pictures (Car, Fish or neutral Face) were shown and the subjects were required to push the button if the two pictures were same. In the Emotion task, thirty-two pairs of pictures (Happy, Angry, Sad or Disgust) were shown and the subjects were required to push the button if the two pictures expressed same emotion. The tasks were counterbalanced across the subjects. SPM12 was used to analyze BOLD fMRI data.
Results: In the Discrimination task, there were no group differences. In the Emotion task, patients with schizophrenia showed reduced BOLD signals in the inferior frontal gyrus only to the disgust faces.
Conclusions: These results suggest that schizophrenia patients have inferior frontal gyrus dysfunction in processing disgust faces. This dysfunction might underlie the paranoid notions in patients with schizophrenia.

Disclosures: **N. Oribe:** None. **H. Kuga:** None. **H. Takao:** None. **S. Fukushima:** None. **Y. Hirano:** None. **T. Onitsuka:** None. **T. Ueno:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.07/WW2

Topic: H.03. Schizophrenia

Title: Tolcapone modulates cortical information processing underlying cognitive control mechanisms in patients with schizophrenia and healthy controls

Authors: *Y. TONG[1], J. A. APUD[2], R. RASETTI[3], J. H. CALLICOTT, III[3], B. KOLACHANA[3], B. K. DAS[3], T. VARGAS[3], B. ZOLTICK[3], D. DICKINSON[3], D. R. WEINBERGER[4], V. S. MATTAY[4], K. F. BERMAN[3]
[1]NIH/NIMH, Bethesda, MD; [2]Office of the Clin. Director, NIH, Natl. Inst. of Mental Hlth., Bethesda, MD; [3]Natl. Inst. of Mental Hlth., Bethesda, MD; [4]Lieber Inst. For Brain Develop., Baltimore, MD

Abstract: Among the multiple cognitive control deficits in schizophrenia, abnormal response inhibition and interference suppression have been frequently reported, and these have been associated with altered dopaminergic signaling in the prefrontal cortex. Converging evidence from animal and human studies suggests a unique role of catechol-O-methyltransferase (COMT) in regulating dopamine in cortical areas, with low activity of the COMT enzyme leading to

improved prefrontal efficiency. In this study we use a modified Flanker paradigm to investigate the potential effect of the COMT inhibitor tolcapone on modulating prefrontal cortical activity related to response inhibition and interference suppression in patients with schizophrenia (SCZs) and healthy controls (NCs).
In this double-blinded, placebo-controlled study, tolcapone was administered orally three times a day for seven days. Participants underwent BOLD fMRI on the seventh day on a GE Signa scanner. The event-related scan session included four conditions with a total of 145 pseudorandomized trials. The participants were asked to indicate the direction of the central arrow on the screen that pointed left or right, while ignoring the flanking stimuli on the side. In the Congruent and Incongruent conditions the flankers were arrows that could be oriented in the same ('congruent') direction or the opposite ('incongruent') direction to the central arrow. The contrast between the Incongruent and Congruent conditions enables us to explore neural processes underlying interference monitoring and suppression. The NoGo condition, which required subjects to withhold their motor response when two pairs of X's flanking the central arrow, served to evaluate response inhibition.
Images were preprocessed in SPM12. Totally 29 SCZs and 62 NCs had good quality fMRI data during both placebo and tolcapone conditions. Both SCZs and NCs showed decreased right IFG activation while on tolcapone when compared to the placebo condition during NoGo trials (pFWE-corr =0.006, right IFG ROI), suggesting improved efficiency in prefrontal cortical information processing during response inhibition. In addition, a significantly greater posterior cingulate cortex (PCC) deactivation was observed during interference monitoring and suppression (incongruent vs. congruent contrast) on tolcapone (pFWE-corr < 0.05, whole brain) in SCZs and NCs. While this finding requires further study, it raises the intriguing possibility that tolcapone in addition to improving efficiency of cortical information processing also improves top down modulation of default mode regions such as PCC during interference detection and suppression.

Disclosures: **Y. Tong:** None. **J.A. Apud:** None. **R. Rasetti:** None. **J.H. Callicott:** None. **B. Kolachana:** None. **B.K. Das:** None. **T. Vargas:** None. **B. Zoltick:** None. **D. Dickinson:** None. **D.R. Weinberger:** None. **V.S. Mattay:** None. **K.F. Berman:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.08/WW3

Topic: H.03. Schizophrenia

Support: NIH Grant T32MH017168

Foerderer Grant from CHOP Research Institute

Title: Ventral hippocampal fMRI hypoactivity and social memory impairment in a 22q11.2 deletion mouse model of schizophrenia

Authors: *J. B. KAHN[1], S. L. WEHRLI[3], S. A. ANDERSON[2,4], D. A. COULTER[1,5,6]
[1]Neurosci., [2]Psychiatry, Univ. of Pennsylvania, Philadelphia, PA; [3]Radiology, [4]Psychiatry, [5]Neurol., [6]Pediatrics, Children's Hosp. of Philadelphia, Philadelphia, PA

Abstract: One in four patients born with the 22q11.2 deletion develops schizophrenia, making this heterozygous deletion the most significant genetic risk factor for the disorder. Both functional magnetic resonance imaging (fMRI) studies in humans and in vivo tetrode recordings in mouse models of the deletion syndrome (DS) have shown abnormal activity patterns in multiple brain regions, including the prefrontal cortex and the hippocampus. To examine neuronal circuits contributing to 22q11.2 phenotypes, we performed gadolinium-enhanced resting state (rs) fMRI in a mouse model of 22q11.2DS (Df(h22q11)/+). While there was no significant difference in blood oxygenation level-dependent (BOLD) signals between the 22q11.2DS mice and controls in the dorsal cortex or caudate putamen, there was a significant decrease in BOLD signals in the ventral hippocampus, consistent with hypofunction. This circuit deficit proved interesting as previous work has found that both patients with 22q11.2DS and those with schizophrenia experience social cognitive deficits, and recent work from the Tonegawa lab has shown that the ventral CA1 is critical for social memory in mice. Based on this potential circuit-dependent/phenotype link, we performed a social memory test on the same fMRI mouse cohort. In the discrimination task, a novel mouse and a familiar mouse are placed in small chambers at opposite ends of an arena. The subject mouse can freely explore the arena, and the amount of time the subject mouse investigates each chamber is quantified. While control animals preferentially explored the novel mouse, the 22q11.2DS mice failed to discriminate between the novel mouse and the familiar mouse, exploring both mice equally. The neural circuit concordance between the rs fMRI and social memory task deficits implicates ventral CA1 as a primary locus for future investigation. Immediate planned steps include bidirectionally manipulating activity in the ventral CA1 via DREADDs (designer receptors exclusively activated by designer drugs) to mimic and/or reverse 22q11.2 phenotypes in control and DS mice, respectively, providing evidence for a causal link between ventral CA1 circuit phenotypes and behavioral deficits in this mouse model of schizophrenia.

Disclosures: J.B. Kahn: None. **S.L. Wehrli:** None. **S.A. Anderson:** None. **D.A. Coulter:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.09/WW4

Topic: H.03. Schizophrenia

Support: DP5 OD012109

Title: Schizophrenia exhibits bi-directional alterations in cortico-striato-cerebellar circuits

Authors: *J. JI[1], C. DIEHL[1], C. SCHLEIFER[1], G. YANG[1], G. CREATURA[1], G. REPOVS[2], J. D. MURRAY[1], A. WINKLER[3], A. ANTICEVIC[1]
[1]Yale Univ., New Haven, CT; [2]Univ. of Ljubljana, Ljubljana, Slovenia; [3]Oxford Univ., Oxford, United Kingdom

Abstract: A key challenge in clinical neuroscience is the identification of robust biomarkers reflecting disease-related alterations in large-scale neural systems. Distributed neural dysconnectivity is considered a hallmark feature of schizophrenia, yet a tension exists between studies pinpointing focal disruptions versus those implicating brain-wide disturbances. Leading theoretical models of schizophrenia have proposed that distributed disruptions in local microcircuits and imbalanced neuronal E-I signaling might underlie pervasive alterations across neural systems, which in turn manifest in specific behaviors. The cerebellum and the striatum communicate reciprocally with the thalamus and cerebral cortex through monosynaptic and polysynaptic connections, forming cortico-striatal-thalamic-cerebellar (CSTC) functional pathways that may be sensitive to brain-wide dysconnectivity in schizophrenia. It remains unknown if the same brain-wide pattern of alterations persists across CSTC systems, or if specific alterations exist along key functional elements of these networks. We characterized whole-brain cerebellar and striatal connectivity using resting-state functional magnetic resonance imaging in 159 chronic schizophrenia patients and 162 matched controls, along each major cerebellar and striatal functional subdivision. Both cerebellar and striatal associative subdivisions revealed consistent brain-wide bi-directional alterations in patients relative to controls, marked by hyper-connectivity with bilateral sensory-motor cortices and hypo-connectivity with association cortex. Furthermore, consistent with previous findings of thalamic dysconnectivity in schizophrenia, these alterations were more pronounced along the executive subdivisions of these systems, suggesting that higher-order functional neural circuits are preferentially disrupted. We additionally validated these results with independent *k*-means clustering of voxelwise striatal and cerebellar between-group connectivity differences. These results implicate a consistent motif of brain-wide alterations in cortico-striato-cerebellar systems in schizophrenia, calling into question accounts of exclusively focal functional connectivity disturbances.

Disclosures: **J. Ji:** None. **C. Diehl:** None. **C. Schleifer:** None. **G. Yang:** None. **G. Creatura:** None. **G. Repovs:** None. **J.D. Murray:** None. **A. Winkler:** None. **A. Anticevic:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.10/WW5

Topic: H.03. Schizophrenia

Title: Role of antipsychotics in disruptions of face detection in schizophrenia

Authors: *P. E. MARINHO[1], N. L. ALMEIDA[2], W. E. M. COMFORT[2], T. M. P. FERNANDES[2], T. L. GOMES[3], N. A. SANTOS[2]
[1]Dept. de Sistemática e Ecologia, [2]Dept. de Psicologia, Univ. Federal Da Paraíba, Joao Pessoa, Brazil; [3]Dept. de Biologia, Univ. Federal Rural de Pernambuco, Recife, Brazil

Abstract: Several studies have reported the effects of antipsychotic medication on visual processing, but there is a lack of studies investigating the relationship between medication use and facial detection. Individuals suffering from schizophrenia often display impairment in their ability to detect emotional facial expressions. Impaired processing of visual information may be one of the key causes of deficits in face detection and can be related to an early detection phase of facial processing. The main purpose of this study was to measure the performance of schizophrenic patients in a face detection task, comparing the effects of typical and atypical antipsychotic drugs. Data were recorded from 33 participants, a group of healthy subjects (n = 11), a group of schizophrenic patients using typical antipsychotics (n = 11) and a group of schizophrenic patients using atypical antipsychotics (n = 11); age range 20-45 years. Patients were recruited from the Psychosocial Support Center in Paraiba, Brazil, diagnosed with schizophrenia according to the Diagnostic and Statistical Manual of Mental Disorders - DSM-V. All subjects were free from any cardiovascular and neurological disorder, identifiable ocular disease and had normal acuity. No abnormalities were detected in the fundoscopic examination and in the optical coherence tomography exam. The stimuli used in the experiment were drawn from the FEI face database. Twelve male and twelve female faces were selected. Detection task aim was to detect the location of a face when presented with a face/non-face pair, using 2-AFC discrimination design. Detection task had a limit of 96 trials. The detection task involved selecting right or left side responses when presented with masked images of face/non-face pairs. The order of test image presentation was determined using Bayesian entropy estimation. The presentation time for each trial was selected to yield the maximum of expected information for prediction of the expected mean threshold. The ocular distance to the screen was set to 80 cm. Performance differed between groups. Schizophrenic patients presented significant deficits in face detection compared to healthy subjects. However, post-hoc analyses suggest that schizophrenic patients using typical antipsychotic presented higher disruption of face detection. These results suggest that not only schizophrenia condition but also the type of antipsychotic

used affected face detection. This highlights the importance of understanding diffuse effects of antipsychotics on visual spatial processing.

Disclosures: P.E. Marinho: None. **N.L. Almeida:** None. **W.E.M. Comfort:** None. **T.M.P. Fernandes:** None. **T.L. Gomes:** None. **N.A. Santos:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.11/WW6

Topic: H.03. Schizophrenia

Support: National Natural Science Foundation of China (No. 31471068, X.W.)

Key Program for International S&T Cooperation Projects of China (MOST, 2016YFE0129100, X.W.)

Thousand Young Talents Program of China (X.W.)

Title: Metacognition dysfunction and disruptions of global balance between brain networks in schizophrenia

Authors: *W. JIA[1], H. ZHU[2], Y. NI[1], J. SU[1], H. JIA[2], X. WAN[1]
[1]State Key Lab. of Cognitive Neurosci. & Learning, Beijing, China; [2]mental illness diagnosis and treatment Beijing Key Lab., Beijing Anding Hosp., Beijing, China

Abstract: Metacognition is referred to as cognition about cognition, a high-level cognitive control process of the forgone cognitive processes. It automatically occur when something goes wrong, and an uncertainty-based “feeling state” is manifest. Metacognition can be separated into two components, the uncertainty monitoring process and the consequent decision revising or controlling process when uncertainty had been detected. Recent studies suggest that metacognition is impaired and it is a core cognitive dysfunction in patients with schizophrenia, but the neural mechanism of the metacognition dysfunction is little understood. Employing a novel metacognition task paradigm - “decision (cognition)-redecision (metacognition)” and functional magnetic resonance imaging (fMRI) technique, we investigated the neural basis of metacognition dysfunction in patients with schizophrenia in the present study. Thirty-eight Schizophrenia (SC) patients and Twenty-one healthy control (HC) subjects were recruited. The participants made two consecutive decisions on the same situation in a perceptual decision-making task (random dot motion, RDM) and judged their confidence levels after decision-making. We found that SC exhibited impairments both in metacognition monitoring and revising processes. Then, we divided SC patients into two groups according to the uncertainty monitoring (UM) ability. The fMRI results showed that the activation in the core nodes of the metacognition

network was significantly reduced in SC, including dorsal anterior cingulate cortex (dACC), which was involved in metacognitive uncertainty monitoring, and the lateral frontopolar cortex (lFPC), which was involved in metacognitive controlling. Furthermore, the anti-correlation between the metacognition network (MCN) and the default mode network (DMN) was alerted in SC. The strength of correlation between the two networks in SC was weaker during uncertainty monitoring, but became stronger in the least-demand task condition, in comparison to those in HC. It appeared that SC failed to properly coordinate the two networks. The group with high UM ability in SC had relatively less DMN suppression, whereas the group with low UM ability in SC had relatively excessive DMN suppression. Most importantly, the SC clinical symptoms were associated with the global balance of the two networks. In conclusion, our current study showed that SC had metacognition dysfunction both in metacognitive monitoring and controlling processes, and this was associated with the disruption of metacognition network, especially the global balance between the metacognition network and the default mode network.

Disclosures: **W. Jia:** None. **H. Zhu:** None. **Y. Ni:** None. **J. Su:** None. **H. Jia:** None. **X. Wan:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.12/WW7

Topic: H.03. Schizophrenia

Support: 503147

508353

Title: Global NMDAR hypofunction increases activity of top-down projections during sensory processing

Authors: ***A. RANSON**, E. BROOM, A. POWELL, J. HALL
Cardiff Univ., Cardiff, United Kingdom

Abstract: Converging evidence implicates NMDAR disruption in the pathogenesis of schizophrenia, a condition in which perceptual disturbances are prominent. Theoretical studies have proposed that perceptual symptoms in schizophrenia might be caused by an increased influence of top-down processing on sensory perception. To explore the cortical circuit changes through which NMDAR hypofunction might cause perceptual symptoms we investigated top-down and bottom-up drive to cortical sensory circuits in awake behaving mice during pharmacologically induced NMDAR hypofunction. Following NMDAR antagonist administration we observed increased activation of anterior cingulate axons providing top-down

input to visual cortex which was coupled with a simultaneous reduction in the primary visual cortex of activity of both PV+ interneurons and putative excitatory neurons and alteration of stimulus encoding. These findings suggest that NMDAR hypofunction may lead to altered perception by modifying the balance of top-down and bottom-up influence on sensory processing.

Disclosures: **A. Ranson:** None. **E. Broom:** None. **A. Powell:** None. **J. Hall:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.13/WW8

Topic: H.03. Schizophrenia

Support: K01MH105615

Title: Neurobiology of reality monitoring: Implications for treatment development in schizophrenia

Authors: *K. SUBRAMANIAM[1], L. B. HINKLEY[1], D. MIZUIRI[1], H. KOTHARE[1], S. VINOGRADOV[2], J. F. HOUDE[1], S. NAGARAJAN[1]
[1]UCSF, San Francisco, CA; [2]Univ. of Minnesota, San Francisco, MN

Abstract: Reality monitoring (RM) is the ability to distinguish the source of internal experiences (self-generated information) from outside reality (externally-derived information). Normally, the healthy brain functions as a continuous comparator prediction engine in which perceiving the world efficiently is fundamentally tied to successfully recognizing one's own self-generated actions - such that intact RM must rely on intact predictive encoding operations. In other words, internal predictions about one's own actions provide reliable self-agency cues; however, if an internal prediction is weak, then external cues from environmental feedback are weighted more so that the action is attributed as being externally-derived. Here, we examine prediction encoding on a low-level vocalization task, as well as how encoding relates to subsequent memory retrieval on a higher-order RM task.
During vocalization, an internal prediction about what one hears when one speaks is compared with the actual auditory feedback of listening to one's speech. Normally, feedback matches prediction encoding, and is registered as self-produced. However, if incoming auditory feedback is perturbed to mismatch predictions (i.e., when participants listen to a perturbed pitch shift while vocalizing), prediction errors are generated. We found that participants respond to the pitch shift by reducing the mismatch between predicted and actual outcome (i.e., by compensating against the pitch shift direction). Interestingly, the more that healthy control participants (HC) compensated against the pitch shift, the less likely they were to identify that information was

self-generated on our RM task. We also found that HC recruit medial prefrontal cortex (mPFC) specifically during encoding of self-generated information, which is also activated during accurate retrieval of self-generated information. By contrast, schizophrenia patients (SZ) have specific self-agency impairments and do not show mPFC activation during self-generated retrieval. These findings indicate that SZ may rely more on environmental externally-derived information, rather than on internal self-generated predictions, to guide RM.
These findings have important therapeutic implications in SZ, suggesting that the more participants rely on environmental feedback, the more they compensate in their pitch, and the less likely they are to correctly identify themselves as the source of self-generated information. In conclusion, pitch compensation may be a useful measure of internal prediction encoding operations that may be used to prime better reality-monitoring (i.e., better self-generated identification) in SZ.

Disclosures: K. Subramaniam: None. **L.B. Hinkley:** None. **D. Mizuiri:** None. **H. Kothare:** None. **S. Vinogradov:** None. **J.F. Houde:** None. **S. Nagarajan:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.14/WW9

Topic: H.03. Schizophrenia

Support: Howard Hughes Medical Institute

Office for Naval Research

UCSD (RO1-MG065571; MH042228; MH079777; MH087889; Brain Behavioral Research Foundation; sidney R. Baer Jr. Foundation)

MSSM (R01-MH065554)

UCLA (R01-MH65707)

PENN (R01-MH65578)

UW (R01-MH65558)

Title: Dynamical clustering of mismatch negativity EEG data in large cohorts of schizophrenia patients and healthy participants reveals functionally distinct subgroups

Authors: *R. KIM[1,4], C. LAINSCSEK[4,5], M. L. THOMAS[2], T. COGS INVESTIGATORS[3], T. J. SEJNOWSKI[4,5], G. A. LIGHT[2]

[1]Neurosci., [2]Psychiatry, [3]UCSD, La Jolla, CA; [4]Computat. Neurobio. Lab., Salk Inst. for Biol. Studies, La Jolla, CA; [5]Inst. for Neural Computation, La Jolla, CA

Abstract: Background
Schizophrenia (SZ), a complex neuropsychiatric disorder, is often referred to as a heterogeneous disorder due to its wide array of clinical symptoms along with many underlying etiologies, most of which are not yet known. It has been suggested that multiple coupled dynamical systems are involved in auditory sensory processing, and the degree to which each component of the coupled systems is affected may contribute to auditory dysfunction present in schizophrenia. By characterizing general dynamical components of brain signals (electroencephalogram; EEG) obtained from patients diagnosed with schizophrenia, important details about the systems-level states associated with auditory information processing deficits can be found.
Methods
A novel clustering algorithm based on delay differential analysis (DDA) was developed to partition EEG signals based on nonlinear dynamical states. The algorithm was then applied to the Consortium on the Genetics of Schizophrenia (COGS-2) EEG recordings. The multi-center COGS-2 dataset, the largest of its kind, contains mismatch negativity EEG recordings from schizophrenia patients (SZ; *n* =877) and non-psychiatric control subjects (NCS; *n* =753).
Results
Our dynamical clustering revealed functionally distinct subgroups (as indicated by P3a and MMN characteristics) within SZ and NCS group. More specifically, we found that (1) a dynamics spectrum exists not only within the SZ group, but also within the NCS group and that (2) a dynamics spectrum might be closely related to a functional/clinical spectrum. These findings suggest that each subgroup identified in NCS and SZ groups displays distinct auditory ERP features (amplitude and latency of MMN and P3a) which might then translate to subgroup-specific psychosocial functioning. Scores from various cognitive test batteries (California Verbal Learning Test-Second Edition, Wechsler Memory Scale, and Penn's Computerized Neurocognitive Battery) will be used to assess cognitive function of each subgroup.
Conclusions
Investigating EEG nonlinear system features associated with impaired auditory information processing in schizophrenia can reveal disorder-specific biomarkers that might help identify individuals at risk, make early diagnosis, and monitor intervention/treatment responses.

Disclosures: R. Kim: None. **C. Lainscsek:** None. **M.L. Thomas:** None. **T. COGS Investigators:** None. **T.J. Sejnowski:** None. **G.A. Light:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.15/WW10

Topic: H.03. Schizophrenia

Support: VA Merit CX000154

NIH R01 MH080187

NIH R01 MH093450

NIH P50MH080272-01

JSPS Grant-in-Aid for Young Scientists B 22791129

JSPS Strategic Young Researcher Overseas Visits Program for Accelerating Brain Circulation S2208

JSPS Grant-in-Aid for Scientific Research C 15K09836

Title: Coincidental disruption of gamma rhythms in first episode schizophrenia

Authors: *Y. HIRANO[1,2], N. ORIBE[1], T. ONITSUKA[1], S. KANBA[1], R. MCCARLEY[3], K. SPENCER[2]
[1]Kyushu Univ., Fukuoka, Japan; [2]VA Boston, Harvard Med. Sch., Boston, MA; [3]VA Boston, Harvard Med. Sch., Brockton, MA

Abstract: BACKGROUND: Deficits in the auditory gamma-band oscillation and volumetric reductions in auditory cortex have been detected shortly after the onset of schizophrenia (SZ), and may be associated with core symptoms such as auditory hallucinations. Disruption of auditory gamma-band oscillation have received considerable interest in this effort, as the basic mechanisms underlying these oscillations are understood and are conserved across species. Evoked gamma oscillation such as the 40-Hz auditory steady-state response (ASSR) are reduced in SZ compared to healthy controls (HC) that could be related to deficits in GABAergic interneuron. Animal models based on NMDA receptor hypofunction demonstrate this deficit as well as an increase in spontaneous gamma power, which has been reported in chronic SZ (Hirano et al.,2015), yet characteristics of these deficits still remain unclear in first-episode schizophrenia (FESZ). Here we examined whether evoked gamma-band ASSR is decreased and spontaneous gamma power is increased in FESZ. **METHODS:** Subjects were 24 FESZ and 38 matched HC. Dipole source localization of dense electrode EEG data was used to examine oscillatory activity in auditory cortices during auditory steady-state stimulation (20/30/40-Hz rates). ICA was used to remove artifacts. Phase locking factor (PLF) and induced power (not phase-locked) were calculated from artifact-free single trial source estimates. Clinical symptoms were assessed by SAPS and SANS. **RESULTS:** Whereas evoked gamma (40-Hz PLF synchrony gamma) was decreased ($p < 0.01$), auditory spontaneous gamma (broadband induced gamma) was increased ($p < 0.01$) during continuous auditory stimuli in FESZ. These two apparently distinctive circuit abnormalities already occurred in the very early stage of the disease. **CONCLUSIONS:** We found coincide disruptions of gamma rhythms, which showed increase in spontaneous gamma (cortical excitability) and decrease in evoked gamma (cortical synchrony failure) during continuous auditory stimuli in FESZ. We propose that assessing ASSR-PLF and spontaneous

gamma in FESZ may provide a sensitive biomarker for the integrity of neural networks that are fundamentally altered in the very early stage of SZ.

Disclosures: Y. Hirano: None. **N. Oribe:** None. **T. Onitsuka:** None. **S. Kanba:** None. **R. McCarley:** None. **K. Spencer:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.16/WW11

Topic: H.03. Schizophrenia

Title: Effects of glutamatergic and dopaminergic drugs on auditory steady state response in common marmosets

Authors: Y. IWAMURA, K. MATSUMOTO, T. NAKAKO, H. IMAI, A. KIYOSHI, T. ENOMOTO, A. MATSUMOTO, M. IKEJIRI, T. NAKAYAMA, Y. OGI, *T. ISHIYAMA, K. IKEDA
Sumitomo Dainippon Pharma Co., Ltd., Osaka, Japan

Abstract: Introduction
Periodic auditory stimulation can elicit the auditory steady state response (ASSR) in the electroencephalogram (EEG) which rapidly entrains to the frequency and phase of the stimulus. ASSRs to 30 and 40 Hz stimulation are reduced in evoked power or phase locking factor (PLF) in patients with schizophrenia (SZ). These deficits have been hypothesized to be associated with N-methyl-D-aspartate receptor (NMDAR) and parvalbumin-positive GABA interneuron abnormalities in SZ. ASSR is expected to be a translational technique to evaluate the drug candidate for SZ. Here we aimed to establish the method of ASSR recording and to investigate the effects of various glutamatergic and dopaminergic drugs on ASSR in common marmosets.
Materials and methods
EEG electrodes were surgically implanted into the skull in six adult marmosets and EEG was recorded from auditory cortex. EEG signals were transmitted wirelessly at a sampling rate of 512 Hz. Auditory clicks at a variable rate (20, 30, 40 and 80 cycles/s; 0.5 s train duration) were generated by Signal software and routed through an auditory processor to a magnetic speaker. Evoked power, induced power and PLF were calculated by wavelet analysis. All drugs were systemically administered before ASSR recording, and the studies were performed in a cross-over manner. All experimental procedures involving animals use were reviewed and approved by the Institutional Animal Care and Use Committee of Sumitomo Dainippon Pharma, Co., Ltd.
Results
We could detect ASSRs (20, 30, 40 and 80 Hz) from common marmoset. NMDAR antagonist (MK-801) induced ASSR deficits with significant reduction of 30 and 40 Hz PLF and evoked

power, which were similarly seen in SZ patients. mGlu2/3 agonist (LY-379268) reduced induced power and increased PLF. D1 agonist (SKF-81297) increased PLF and evoked power. On the other hand, D1 receptor antagonist (SCH-39166) decreased PLF and evoked power. D2 agonist (quinpirole) did not change PLF, but increased evoked and induced power. D2 antagonist (raclopride) decreased PLF and evoked power. Antipsychotic (risperidone) did not increase PLF.

Conclusion

We could successfully measure ASSR from marmosets and demonstrated that NMDAR antagonist induced SZ-like ASSR deficits such as reduction of PLF and evoked power. Moreover, we showed that increase of both ASSR parameters are induced by D1 agonist, which is reported to enhance cognitive function in marmoset, but not by the other drugs tested. Thus, ASSR in marmoset is considered to be a translational biomarker of glutamatergic and dopaminergic neurotransmission, which may be involved at least in part in cognitive function.

Disclosures: **Y. Iwamura:** None. **K. Matsumoto:** None. **T. Nakako:** None. **H. Imai:** None. **A. Kiyoshi:** None. **T. Enomoto:** None. **A. Matsumoto:** None. **M. Ikejiri:** None. **T. Nakayama:** None. **Y. Ogi:** None. **T. Ishiyama:** None. **K. Ikeda:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.17/WW12

Topic: H.03. Schizophrenia

Support: NIH Grant R01MH101102

Title: Abnormal hippocampal-mPFC synchrony in the KCNH2-3.1 transgenic mouse model

Authors: ***M. REN**[1], J. ZHU[1], Y. LI[1,2], S. ZHU[1,2], K. MEZEIVTCH[1], Z. HU[1], S. QIN[1], X. LI[3], Q. TIAN[1], D. PARADES[1], Q. CHEN[1], K. H. WANG[3], D. R. WEINBERGER[1], F. YANG[1]
[1]The Lieber Inst. for Brain Develop., Baltimore, MD; [2]Chongqing Med. Univ., Chongqing, China; [3]Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: The KCNH2-1A potassium channel conducts delayed-rectifier potassium currents that have a rapid activation and relatively slow inactivation in deactivation kinetics. Our previous work has identified a novel primate isoform of KCNH2-1A channel, the KCNH2-3.1 potassium channel, in human brain associated with schizophrenia. And our studies also showed that KCNH2-3.1 transgenic mice exhibit alterations in neuronal structure and microcircuit function in the hippocampus and prefrontal cortex, areas affected in schizophrenia. Neuronal oscillations have been hypothesized to play an important role in cognition and its ensuing behavior, but evidence that links a specific neuronal oscillation to a discrete cognitive event is largely unknown. It has been proposed that failure of functional integration and flow of information

between hippocampus and mPFC results in cognitive deficits in schizophrenia patients. The possibility of functional impairments between hippocampus and mPFC as key pathophysiological mechanisms associated with schizophrenic phenotypes in KCNH2-3.1 mice has not been explored before. In this study, to investigate functional connectivity in these mice, we measured the synchronization of neural activity between the hippocampus and the prefrontal cortex during the performance of a delayed nonmatch-to-place (DNMP) T maze task, which is analogous to a cognitive function disrupted in the schizophrenia. KCNH2-3.1 mice, which display significantly impaired learning compared to control mice in the DNMP T maze task, showed robustly reduced synchrony measured by coherence of local field potentials in ventral hippocampus and mPFC, particularly in beta and gamma oscillations. Interestingly, higher beta and gamma oscillation power is observed in KCNH2-3.1 transgenic mice. These findings provide further insights into the precise alterations in hippocampal-mPFC connectivity observed in this schizophrenia-associated animal model, and could ultimately help elucidate the molecular and cellular basis underlying schizophrenia pathogenesis.

Disclosures: **M. Ren:** None. **J. Zhu:** None. **Y. Li:** None. **S. Zhu:** None. **K. Mezeivtch:** None. **Z. Hu:** None. **S. Qin:** None. **X. Li:** None. **Q. tian:** None. **D. Parades:** None. **Q. chen:** None. **K.H. Wang:** None. **D.R. Weinberger:** None. **F. Yang:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.18/WW13

Topic: H.03. Schizophrenia

Support: NIMH Grant MH090067

Postdoctoral Fellowship SALSI

Title: The orexin receptor antagonist TCS 1102 reverses aberrant dopamine system function in a rodent model of schizophrenia

Authors: *S. M. PEREZ[1], M. PATTON[1], D. J. LODGE[2]
[1]Pharmacol., [2]UTHSCSA, San Antonio, TX

Abstract: Aberrant regulation of dopamine system function is thought to contribute to psychosis in schizophrenia patients; however, the brain regions associated with this dysregulation have not been conclusively demonstrated. We have recently demonstrated that medium spiny neurons in the nucleus accumbens (NAc) receive convergent input from the ventral hippocampus (vHipp) and paraventricular nucleus of the thalamus (PVT). Furthermore, inactivation of either the vHipp or PVT is sufficient to reverse aberrant dopamine system function in rodent models of

schizophrenia. Using, chemogenetic experiments we now provide conclusive evidence that the thalamic input to the NAc plays a role in the regulation of dopamine neuron activity. These data demonstrate that the vHipp and thalamus (specifically the PVT) work in concert to regulate VTA dopamine neuron population activity. Such data are important as they provide evidence that thalamic abnormalities may contribute to the aberrant dopamine system function observed in schizophrenia and suggest that the PVT may be a novel site for intervention in psychosis. To examine this, we explored the orexin system, which is known to provide a dense innervation of the PVT. Indeed, we now show that orexin receptors are expressed on PVT neurons projecting to the NAc and may serve as a substrate for pharmacological manipulation of this pathway. Here, we provide evidence that both systemic and intracranial (PVT) administration of the orexin receptor antagonist, TCS1102, can normalize aberrant dopamine system function in a rodent model of schizophrenia. Collectively, these data suggest that targeting orexin signaling in the thalamus may represent a novel site of intervention for psychosis associated with schizophrenia.

Disclosures: S.M. Perez: None. **M. Patton:** None. **D.J. Lodge:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.19/WW14

Topic: H.03. Schizophrenia

Support: CNPq - Brazil

CAPES - Brazil

Title: Intra-nucleus accumbens infusion of quinpirole recovers mesolimbic hyperdopaminergia induced by systemic administration of sulpiride

Authors: *S. B. DE SOUZA[1], E. MCKIMM[3], S. M. TRABOSH[3], C. D. BLAHA[2], C. DA CUNHA[4]
[1]Neural Engin. Labs., [2]Dept. of Neurologic Surgery, Mayo Clin., Rochester, MN; [3]Dept. of Psychology, Univ. of Memphis, Memphis, TN; [4]Univ. Federal do Parana, Curitiba, Brazil

Abstract: Dysfunctional hyperdopaminergic signaling, caused by increased release of dopamine from presynaptic terminals, in the mesolimbic system is associated with psychotic symptoms in schizophrenic patients. Dopamine D2 receptors (D2R) antagonists are clinically used to control the manifestation of such symptoms, however, chronic administration has shown to induce supersensitivity of postsynaptic D2 receptors. Yet, both pre and postsynaptic D2R can be occupied by D2R antagonists which contributes to the complexity of the treatment. The goal of the present study was to elucidate the effects of low (presynaptic) and high (pre- and

postsynaptic) doses of the D2R antagonist sulpiride in the evoked dopamine release controlled by D2 autoreceptors in the nucleus accumbens (NAc). We used fixed potential amperometry with the application of low frequency electrical conditioning stimuli (CS = 5, 10, 20 and 40 prepulses at 15Hz, 0.5ms/phase, 0.8mA) to the medial forebrain bundle of DBA/2J mice to evaluate D2 autoreceptors blockade. By increasing the number of CS, we observed the inhibition of dopamine release evoked by a test electrical stimulation (10 pulses at 50Hz, 0.5ms, 0.8mA) applied 0.3s after offset of the CS when compared to a control stimulus of the same magnitude. Intraperitoneal injection of high dose sulpiride blocked both the inhibition of dopamine release controlled by D2 autoreceptors and the dopamine uptake, mediated by the dopamine transporter (DAT). However, a low dose of sulpiride only blocked the inhibition of dopamine release when a small amount of extracellular dopamine was previously released by 5 prepulses CS. Next, intra-NAc infusion of the D2/D3 agonist quinpirole reversed the inhibition blockade effect of both doses of sulpiride and amplified the dopamine uptake previously disrupted by the high dose of sulpiride. Our data shows that mesolimbic hyperdopaminergia is induced by administration of high doses of D2R antagonists through a mechanism mediated by the blockade of D2 autoreceptors and DAT. In addition, we show that balanced activation of D2/D3 receptors is important to maintain physiological mesolimbic dopamine levels. These results further suggest that choosing a pharmacological agent capable of regulating dopaminergic levels in the brain can be a better approach to treat psychotic symptoms experienced by schizophrenic patients.

Disclosures: S.B. De Souza: None. **E. McKimm:** None. **S.M. Trabosh:** None. **C.D. Blaha:** None. **C. Da Cunha:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.20/WW15

Topic: H.03. Schizophrenia

Support: Consejo Nacional de Ciencia y Tecnologia PN 2016-01-465

Consejo Nacional de Ciencia y TecnologiaI NFR-280414

Red Temática Células Troncales y Medicina Regenerativa (No. 271609)

Title: Focal cortico-striatal tract deymelination in juvenile mice promotes neuro-functional changes related to social and motor-stereotypical behaviours

Authors: *G. MENDEZ-VICTORIANO[1], R. E. REYNA-GUTIERREZ[1], J. M. VEGA-RIQUER[2,1], N. MOY-LOPEZ[1], J. GUZMAN-MUNIZ[1], O. GONZALEZ-PEREZ[1]

[1]Sch. of Psychology/Neurosciences Lab., Univ. of Colima, Colima, Mexico; [2]Med. Sci. PhD Program, Sch. of Medicine/University of Colima, Colima, Mexico

Abstract: Introduction: Recent evidence suggest that schizophrenia's pathogenesis involves white matter dysregulations, cortical hypofunction, striatal hyper-function, and neuroanatomical alterations in brain areas related to social and motor-stereotypical behaviors. During adolescence, white matter alterations that affect neuronal activity have been related to the symptomatic appearance of this disorder. Schizophrenia-like animal models that involve demyelination have been standardized to elucidate the possible relationship between myelin affection with behavioral/anatomical brain alterations. However, these models produce a diffuse white matter affection and cannot establish a possible anatomical and functional relationship. **Purpose:** to evaluate the effects of focal cortico-striatal tract demyelination on neuronal activity, social and motor-stereotypical behavior. **Method:** 20 Six-week-old CD1 mice were injected bilaterally with 0.1% Lysolecithin (experimental) or 0.9% saline solution (control) at +2.3 mm, +/- 1.0 mm, -1.5 mm coordinates related to Bregma. Six days after surgery, to evaluate social and motor-stereotypical behaviors, we used two behavioral tests: the three-chamber sociability test and the marble burying test. Neuronal activity was determined by counting the number of c-Fos+ cells in prefrontal cortex and caudoputamen. **Results:** The demyelinated group showed a decrease in the number of marbles buried and in social motivation and social novelty as compared to the control group. We found an increase in the number of c-Fos+ cells in caudoputamen and a decrease in prefrontal cortex of the demyelinated group when compared to the control group. **Conclusions:** Focal cortico-striatal tract demyelination promotes neuroanatomical changes related to social and motor-stereotypical behaviors. These findings suggest that lysolecithin injection is a suitable model to evaluate the relationship between brain regions demyelination and behavior in some neurodevelopmental disorders.

Disclosures: G. Mendez-Victoriano: None. **R.E. Reyna-Gutierrez:** None. **J.M. Vega-Riquer:** None. **N. Moy-Lopez:** None. **J. Guzman-Muniz:** None. **O. Gonzalez-Perez:** None.

Poster

435. Human and Animal Studies of Circuits and Systems in Schizophrenia

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 435.21/WW16

Topic: H.03. Schizophrenia

Support: NIH R21 MH103541

Title: A study of auditory cortical cross-frequency coupling in schizophrenia

Authors: *N. RAMAKRISHNAN, N. R. MURPHY, C. P. WALKER, N. R. POLIZZOTTO, D. P. WOMACK, R. Y. CHO
Psychiatry, Univ. of Texas Hlth. Sci. Ctr., Houston, TX

Abstract: Background: Abnormal neural oscillatory dynamics in schizophrenia patients are thought to give rise to cognitive deficits in the illness. These include gamma oscillatory disturbances as well as possible disturbances in cross-frequency coupling (CFC), the modulation of high-frequency oscillations by lower frequency oscillations. The aim of this study was to investigate phase-amplitude CFC in the auditory cortex during 40 Hz steady-state stimulation and their potential disturbance in schizophrenia patients. **Methods:** Schizophrenia patients (n=10) and healthy controls (n = 9) were presented with a 40 Hz auditory click train stimuli (1000 ms) during high-density (305 channel) Magnetoencephalography (MEG) recording. The MNE Python toolbox was used to process the raw MEG signals and MATLAB scripts were used to compute sensor-level spectral and CFC analysis of the 40 Hz auditory steady state response (ASSR). We defined a functional region of interest (ROI) using 40hz frequency power response over the course of the entrainment period (0 to 1000ms), and averaged over sensors selected within the top quartile. The co-modulogram for CFC was examined for phases of lower frequency oscillations ranging 2 to 16 Hz amplitudes of higher frequency oscillations ranging 20-60 Hz using the mutual information as a measure of interaction between frequency bands. CFC during the entrainment period was normalized by subtracting CFC during the baseline period. **Results:** Steady state gamma power was not significantly reduced in patients relative to controls. Estimates of CFC, however, demonstrated disparities in delta-gamma CFC, with healthy controls exhibiting higher coupling compared to schizophrenia patients. **Conclusions:** This is the first MEG study to investigate cross-frequency neuronal interactions during the auditory steady state responses in schizophrenia patients. Despite comparable gamma oscillatory power, CFC appears to be impaired in patients. Thus, while there may exist relative preservation of the ability to entrain activity to high frequency stimuli, the modulation of such activity by lower frequency activity may be diminished in the illness.

Disclosures: N. Ramakrishnan: None. **N.R. Murphy:** None. **C.P. Walker:** None. **N.R. Polizzotto:** None. **D.P. Womack:** None. **R.Y. Cho:** None.

Poster

436. Staining and Imaging Techniques

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 436.01/WW17

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Title: Quantification of immunohistochemistry on adjacent sections comparing fluorescent and DAB markers

Authors: *B. TIPTON, J. BAUN, T. YORK, C. ZURHELLEN, R. C. SWITZER, III
Lab., Neurosci. Associates, Knoxville, TN

Abstract: The location of antibody binding sites in brain tissue sections is commonly detected with fluorescent markers or with colored reaction products such as diaminobenzidine. We sought to answer the question: Do both types of detection schemes reveal the antibody binding sites to the same extent as judged by density of staining?
Adjacent sections of tissue were stained with several antibodies, including GFAP for astrocytes and Iba1 for microglia from rat brains that were unilaterally rendered ischemic by middle cerebral artery occlusion. Sets of adjacent sections were immunohistochemically stained free-floating using standard IHC methods. For one set, the antibody binding site was detected using a secondary antibody conjugated with a fluorescent molecule. The second set was stained using the sequence of a secondary antibody conjugated with a biotin molecule, followed by an avidin-HRP complex and then reacting with diaminobenzidine and H_2O_2.
Two areas of interest were digitally captured for each detection scheme/stain: An ischemic area in one hemisphere and an unaffected area in the other hemisphere. The same region of interest for each stain was analyzed on adjacent sections to rule out any random differences in brain areas. Images of both the DAB and fluorescent stains were converted to 8-bit grayscale. The 8-bit grayscale images of fluorescent-stained tissue were inverted to mimic the 8-bit grayscale images of the DAB images. All 8-bit grayscale images were converted to a binary image for densitometric analysis. Densitometry (measured as the percent area occupied by signal) was performed to determine any differences between the DAB and fluorescent images. The percent area in both the ischemic and normal areas of interest were analyzed in both DAB and fluorescent images for each stain.
There were dramatic differences observed between the two staining methods. In both the GFAP and Iba1 stains there were obvious variations in staining intensity in both the affected and normal brain regions. This is not surprising as there is considerably greater amplification of the antibody presence using the DAB multiple stage sequence vs. the “single” stage fluorophore-secondary antibody sequence. Nonetheless, the degree of hypertrophied astrocytes and microglia seen with the fluorescent method is meager compared to the images using the DAB sequence.

Disclosures: B. Tipton: None. **J. Baun:** None. **T. York:** None. **C. Zurhellen:** None. **R.C. Switzer:** None.

Poster

436. Staining and Imaging Techniques

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 436.02/WW18

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Title: Improved application of the electrophoretic tissue clearing technology, CLARITY, to intact solid organs

Authors: *S. PARK[1], H. LEE[2]
[1]Predictive Model Res. Ctr., Korea Inst. of Toxicology, Daejeon, Korea, Republic of; [2]Keimyung Univ. Sch. of Med., Dalseo-Gu, Korea, Republic of

Abstract: Mapping of tissue structure at the cellular, circuit, and organ-wide scale is important for understanding physiological and biological functions. A bio-electrochemical technique known as CLARITY used for three-dimensional anatomical and phenotypical mapping within transparent intact tissues has been recently developed. This method provided a major advance in understanding the structure-function relationships in circuits of the nervous system and organs by using whole-body clearing. Thus, in the present study, we aimed to improve the original CLARITY procedure and developed specific CLARITY protocols for various intact organs. We determined the optimal conditions for reducing bubble formation, discoloration, and depositing of black particles on the surface of tissue, which allowed production of clearer organ images. We also determined the appropriate replacement cycles of clearing solution for each type of organ, and convincingly demonstrated that 250–280 mA is the ideal range of electrical current for tissue clearing. We then acquired each type of cleared organs including brain, pancreas, liver, lung, kidney, and intestine. Additionally, we determined the images of axon fibers of hippocampal region, the Purkinje layer of cerebellum, and vessels and cellular nuclei of pancreas. CLARITY is an innovative biochemical technology for the structural and molecular analysis of various types of tissue. We developed improved CLARITY methods for clearing of the brain, pancreas, lung, intestine, liver, and kidney, and identified the appropriate experimental conditions for clearing of each specific tissue type. These optimized methods will be useful for the application of CLARITY to various types of organs.

Disclosures: S. Park: None. **H. Lee:** None.

Poster

436. Staining and Imaging Techniques

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 436.03/WW19

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: TUBA- Turkish Academy Of Sciences

Hacettepe University, Institute of Neurological Sciences and Psychiatry

Title: Fluorescent biomarkers as reporters of poor quality transcardial perfusion-fixation in frozen brain sections: A much needed tool for experimental neuroscience

Authors: *A. DEHGHANI[1], *A. DEHGHANI[1], H. KARATAS KURSUN[1], A. CAN[2], M. YEMISCI OZKAN[1,3], E. EREN KOCAK[1,4], T. DALKARA[1,3]
[1]Hacettepe Uni. Inst. Neurol. Sci. and Psych., Ankara, Turkey; [2]Ankara Univ. Sch. of Med., Dept. of Histology and Embryology, Ankara 06100, Turkey; [3]Hacettepe University, Fac. of medicine, Dept. of Neurol., Ankara, Turkey; [4]Hacettepe University, Fac. of Medicine, Dept. of Psychiatry, Ankara, Turkey

Abstract: Fixation via transcardial perfusion is performed for preserving the tissue uniformly in a life-like state without allowing any hypoxia/hypoperfusion-induced alterations during sacrificing animals. Since frozen sections are commonly used in studies with fluorescent-tagged antibodies, co-localization imaging and transgenic animals expressing fluorescent proteins, we need tools to detect poor perfusion in frozen sections to ensure that observed changes are genuine but not induced by non-optimal tissue fixation. Adult naive Swiss albino mice were anesthetized with 1 mg/g chloral hydrate (i.p.). Transcardial perfusion parameters were varied by changing the flow rate of heparinized saline and 4% paraformaldehyde (PFA) as well as perfusion duration. Brains were extracted and prepared for Nissl, Hematoxylin/Eosin (H&E), Hoechst-33258, YOYO-1, TO-PRO3 staining or NeuN and HMGB1 immunolabeling. Images were taken with fluorescent, phase-contrast, differential interference contrast (DIC), laser scanning confocal and scanning electron microscopy (SEM). The images taken from cortical and subcortical regions of 8 well-perfused brains and 10 poor-perfused brains were quantified at 20x. The nuclear diameter was measured in Nissl at 40x. All fluorescent stainings of frozen sections including Hoechst, YOYO-1, TO-PRO3, NeuN and HMGB1 exhibited a significant number of dysmorphic neuronal nuclei resembling donats (nuclei with unstained core) in poor-perfused brains. In Nissl or H&E-stained sections, these neurons displayed nuclear swelling associated with perineuronal astrocyte endfeet swelling, classical histopathologic hallmarks of early hypoxic/ischemic injury. The average ratio of donat-shaped neurons to Hoechst-labeled nuclei was less than 5% in well-fixed PFA-perfused mice. In addition to studies co-localizing Nissl and fluorescent staining performed with phase-contrast and DIC confocal microscopy, SEM images also clearly showed loss of tissue integrity in poor-perfused brains. Translocation of HMGB1 from nucleus to cytoplasm in these dysmorphic neurons also confirmed that they were exposed to hypoxic/ischemic stress during perfusion-fixation. These findings suggest that dysmorphic 'donat-shaped' neuronal nuclei in Hoechst, TO-PRO3, YOYO-1 stainings as well as NeuN and HMGB1 immunolabeling can be regarded as histological biomarkers reporting poor quality of transcardial perfusion-fixation in frozen sections. Hoechst staining, which is an easily applicable and widely used nuclear marker can be employed for assessing the quality of perfusion-fixation by taking into account the 5% threshold.

Disclosures: A. Dehghani: None. **H. Karatas Kursun:** None. **A. Can:** None. **M. Yemisci Ozkan:** None. **E. Eren Kocak:** None. **T. Dalkara:** None.

Poster

436. Staining and Imaging Techniques

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 436.04/WW20

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant R01 CA211861

NIH Grant R21 AR066931

NIH Grant R21 EB022788

Title: Ultra-luminous labels for imaging of individual molecular targets in brain cells

Authors: *V. V. DIDENKO
Neurosurg., Baylor Col. of Med., Houston, TX

Abstract: We describe new highly luminous fluorescent labels developed for biomedical imaging of individual, low abundance molecular targets in brain cells. The new ultra-luminous fluorescent tags are the rod-shaped nanoparticles assembled from organic polymers and nanotubes. They can be individually observed under the fluorescence microscope. These composition fluorophores are significantly lighter and less dense as compared to Q-dots. Such properties ensure their low pull forces in repetitive washing. When conjugated to antibodies these ultra-bright labels make possible the routine detection and observation of individual loosely attached or fragile molecular targets. They can be advantageous for *in situ* labeling of exosomes, microvesicles and individual DNA breaks in brain cells. The ultra-thin profile of the new florescent labels is also beneficial in multiplex labeling of tissue sections by reducing steric hindrance during detection of the closely located molecular targets.

Disclosures: V.V. Didenko: None.

Poster

436. Staining and Imaging Techniques

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 436.05/WW21

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant P41GM103412

NIH Grant GM086197

Branfman Family Foundation

Title: Split-miniSOG for imaging intracellular protein-protein interactions by correlated light and electron microscopy

Authors: *D. BOASSA[1], S. R. ADAMS[2], S. PALIDA[2], V. LEV-RAM[2], J. HU[1], Q. XIONG[2], S. PHAN[1], M. ELLISMAN[1], J. T. NGO[3]
[1]CRBS, NCMIR, [2]Pharmacol., Univ. of California San Diego, La Jolla, CA; [3]Biomed. Engin., Boston Univ., Boston, MA

Abstract: We have engineered a protein fragment complementation probe for imaging intracellular protein-protein interactions by electron microscopy (EM). The probe was constructed by bisection of miniSOG, a fluorescent flavoprotein derived from the LOV2 domain of *Arabidopsis* phototropin. When brought together by interacting proteins, miniSOG fragments reconstitute a functional reporter, permitting labeled complexes to be visualized by fluorescence detection as well as by EM imaging through miniSOG-mediated photooxidation of 3-3'-diaminobenzidine (DAB). The protein fragments are relatively small (10.96 and 5.15 kDa), and interact with one another reversibly, which should minimize perturbation of the native dynamics of the tagged interacting partners. We describe the use of split-miniSOG to the visualization of protein aggregates associated with neurodegenerative diseases, including the homomeric aggregation of alpha-synuclein (AS). In neurons, AS is expressed as both a soluble and membrane-associated protein and is the main component of Lewy bodies, the cellular aggregates seen in the brains of patients with PD and other neurodegenerative conditions. Use of split-miniSOG to detect AS-aggregates provides distinct advantages, such as the ability to distinguish between soluble and aggregated forms of the disease-associated molecule. We have exploited this advantage and generated AS chimeras tagged with complementary miniSOG fragments. Labeled protein aggregates were detected as darkly stained inclusions in the cell bodies and dendrites of transfected neurons, formations consistent with "Lewy bodies" and "Lewy dendrites." Using this technology we are able to locate both fibrillar and oligomeric forms of AS within neuronal cells with unprecedented resolution, precision, and sensitivity.

Disclosures: D. Boassa: None. **S.R. Adams:** None. **S. Palida:** None. **V. Lev-Ram:** None. **J. Hu:** None. **Q. Xiong:** None. **S. Phan:** None. **M. Ellisman:** None. **J.T. Ngo:** None.

Poster

436. Staining and Imaging Techniques

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 436.06/WW22

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: CIHR 148882

Title: Direct observation of local protein synthesis in single cells *In vivo*

Authors: *I. KAYS[1], B. E. CHEN[2]
[1]McGill Univ. Hlth. Ctr., Montreal, QC, Canada; [2]Ctr. for Res. in Neurosci., McGill Univ., Montreal, QC, Canada

Abstract: Proteins are the end products of gene expression and the executors of cell function. Detecting when and how much a protein molecule is synthesized is important for understanding cell function, but current methods have poor cellular or temporal resolution or are destructive to cells. Here, we developed a technique to detect and quantify subcellular protein synthesis events at millisecond resolution *in vivo*. This Protein Translation Reporting (PTR) technique uses a genetic tag that produces a stoichiometric ratio of a small peptide portion of a split fluorescent protein and the protein of interest during protein synthesis. We show that the split fluorescent protein peptide can generate fluorescence within milliseconds upon binding the larger portion of the fluorescent protein, and that the fluorescence intensity is directly proportional to the number of molecules of the protein of interest synthesized. We split a green and red fluorescent protein to allow for detection of synthesis of each parental allele of a gene in two channels to examine the allele-specific distribution of protein synthesis. We also split a photoswitchable fluorescent protein to photoconvert the reconstituted fluorescent protein to a different color, to continually and arbitrarily reset the time of detection of synthesis events. Using PTR, we directly observed and quantified protein synthesis events subcellularly over time *in vivo*.

Disclosures: I. Kays: None. **B.E. Chen:** None.

Poster

436. Staining and Imaging Techniques

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 436.07/WW23

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: A.P. Giannini Postdoctoral Fellowship

Beckman Technology Innovation Grant

Title: Nucleomic substrates for social behavior in GnRH1 neurons in an African cichlid fish

Authors: *S. ALVARADO, R. A. FERNALD
Biol., Stanford Univ., Palo Alto, CA

Abstract: Changing behavior to adapt to novel situations is critical for surviving dynamic environments. In this respect, genes are important constituents of behavior but are static and cannot explain the plasticity observed in behavioral phenotypes. Epigenetic mechanisms, such as histone modifications or DNA methylation, lend plasticity to genes that determine behavior with reversible modification. While molecular tools exist that can shed light on epigenetic modifications to whole tissues, few approaches offer insight to individual nuclei. The development of array tomography provides the power to resolve microstructures such as the synapse and nuclei within neurons to 50-200 nm resolution through resin embedding and serial sectioning. We use a fish model system, Astatotilapia burtoni, which has dramatic behavioral polyphenism between dominant and non-dominant males that produce concomitant morphological, neuroanatomical, and molecular changes. Social status in these animals is accompanied with a repertoire of molecular changes that are localized within specific cell populations in the brain. Specifically, dominant males show enlarged soma within their GnRH1 neurons compared to their subordinate counterparts. In this study, we employed the use of a suite of chromatin immunoprecipitation-validated antibodies to trace the 3D organization of chromatin with array tomography in single GnRH1 neurons. In addition to immunostaining histone modifications, we paired our approach with fluorescent in situ hybridization of tiled array generated probes to label genes within active and inactive chromatin marks. As epigenetic mechanisms are emerging as novel mediators of molecular pathologies in general, this approach can be of critical importance for neurosciences and molecular approaches in many other fields.

Disclosures: S. Alvarado: A. Employment/Salary (full or part-time):; Stanford University. F. Consulting Fees (e.g., advisory boards); Thwacke LLC. **R.A. Fernald:** A. Employment/Salary (full or part-time):; Stanford University.

Poster

436. Staining and Imaging Techniques

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 436.08/WW24

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: KAKENHI Grant 15K10377

KAKENHI Grant 15K19196

Title: Study on antioxidant measurement in human brain using MRS

Authors: *T. MURASE, M. UMEDA, T. HIGUCHI
Meiji Univ. of Integrative Med., Kyoto, Japan

Abstract: [Introduction] Glutathione (GSH) and ascorbic acid (Asc, vitamin C) are the most concentrated chemical antioxidants in the central nervous system. Measuring GSH and Asc concentrations non-invasively in vivo is very important in investigating the role of this antioxidant in normal and diseased brain functions. Proton MR spectroscopy (1H-MRS), which can be measured using clinically popular MRI scanner, can detect non-invasively comprehensive metabolites of the human brain. However, it is impossible to reliably quantify the full range of 1H-MRS detectable metabolites in vivo, due to the spectral overlap of resonances from different compounds observed at low magnetic field strength. Since GSH and Asc are metabolites with perfect spectral overlap with other metabolites, "spectrum editing" method is essential. Therefore, in this study, we investigated whether phantom and human brain can measure GSH and Asc by clinical 3 TMRI using spectrum editing method.
[Methods] GSH phantom (10mM GSH and 10mM NAA) and Asc phantom (10mM Asc and 10mM NAA) were acquired at physiological pH. Five healty volunteers gave informed consent for this study. The experimental equipment consisted of a 3T MRI device (SIEMENS company, Trio A Tim system) and a 32-channel head coil. Single voxel MRS (MEGA-PRESS) was used to measure GSH levels for phantom and human brain with the following parameters: TR/TE = 3,000/68 ms, NEX = 128 (*2), VOI = 20mm *30mm *20mm, choice excitation pulse = 4.56 ppm (editing on) / 7.5ppm (editing off). MEGA-PRESS was used to measure Asc levels for phantom and human with the following parameters:The measurement parameters of Asc editing single voxel MRS (MEGA-PRESS) is TR/TE = 3,000/160 ms, NEX = 128 (*2), VOI = 20mm *30mm *20mm, choice excitation pulse =4.01 ppm (editing on) / 5.39ppm (editing off). The spectral signals of GSH and Asc are obtained by the difference between editing off and editing on data. MRS data analysis used 1D FT-NMR (ISBN: 9784782705971).
[Results & Discussion] As a result of phantom experiment, it was confirmed that GSH and Asc can measure 3T MRI scanner using MEGA-PRESS. On the other hand, GSH signals were observed in human brain experiments, but Asc signals could not be observed. The previous studies using a high magnetic field MRI apparatus have reported that GSH and Asc are present in the human brain at a concentration of about 1 mM. Asc was not observed in the human brain, because it seems that optimization of measurement conditions is insufficient, we will investigate in the future.

Disclosures: **T. Murase:** None. **M. Umeda:** None. **T. Higuchi:** None.

Poster

436. Staining and Imaging Techniques

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 436.09/WW25

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: BRAIN Initiative award from NIH/NIMH (U01MH106008)

Title: Novel knockin mouse lines for Cre-depenent sparse labeling of genetically-defined, single neurons for brainwide morphological analyses

Authors: *M. B. VELDMAN[1], N. N. FOSTER[2], M. ZHU[2], M. BECERRA[2], T. L. DAIGLE[3], H. ZENG[4], H. DONG[2], X. W. YANG[1]
[1]Semel Inst., UCLA, Los Angeles, CA; [2]USC Stevens Neuroimaging and Informatics Inst., Keck Sch. of Med. of USC, Los Angeles, CA; [3]Allen Inst. For Brain Sci., Seattle, WA; [4]Structured Sci., Allen Inst. for Brain Sci., Seattle, WA

Abstract: Detailed analyses of neuronal cell morphology in the brain often requires methods (such as Golgi stain) to sparsely and completely label the neuronal cell body as well as their dendritic and axonal processes. To achieve genetically-directed sparse neuronal labeling, our laboratory has provided proof-of-concept that D1 dopamine receptor BAC driven unstable repeat can undergo frameshift to sparsely and stochastically switch on reporter gene expression in a subset of D1-expressing neurons (Lu, 2016). To demonstrate the general utility of this method (called MORF) for labeling many other neuronal (and non-neuronal) cell types, and to achieve more complete labeling of the neuronal processes, we developed here Cre-dependent MORF reporter knockin mouse lines. By crossing one of the lines to a ubiquitous Cre or neuronal cell-type-specific Cre lines, we achieved Cre-dependent, sparse and stochastic labeling of distinct cell populations (e.g. D2 medium spiny neurons or cerebellar Purkinje cells) in the mouse brain. We will describe the distinct reporters used in our knockin MORF lines, their labeling frequencies upon Cre induction, and their utility in brainwide analyses of genetically-defined single neuronal morphology. Future application of such Cre-dependent MORF mice may facilitate the study, at single neuron resolution, the development, function, dysfunction and degeneration of mammalian neurons in vivo.

Disclosures: M.B. Veldman: None. **N.N. Foster:** None. **M. Zhu:** None. **M. Becerra:** None. **T.L. Daigle:** None. **H. Zeng:** None. **H. Dong:** None. **X.W. Yang:** None.

Poster

436. Staining and Imaging Techniques

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 436.10/WW26

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: Canadian Institutes of Health Research (148882)

Title: Quantification of allele-specific protein expression over time in single cells *In vivo*

Authors: *C.-A. LO[1], B. E. CHEN[2]
[1]Ctr. for Res. In Neurosci., Montreal, QC, Canada; [2]Ctr. for Res. in Neurosci., McGill Univ., Montreal, QC, Canada

Abstract: Protein production is a dynamic and regulated process in cells. However, little is known about the dynamics of protein synthesis in single cells in an animal. Here, we use a fluorescence based non-invasive protein quantification technique to tack protein production dynamics over time in single cells in animals. Specifically, we used a genetic tag, called a protein quantitation reporter (PQR), connecting a gene of interest to a fluorescent reporter gene. Protein quantification is based on the equimolar production of the protein of interest and the fluorescent reporter from the same genomic locus, but the fluorescent reporter is separated from the protein of interest during protein synthesis. Therefore, the fluorescent output of a cell (i.e., its brightness) is linearly proportional to the number of molecules of the protein of interest produced. To monitor endogenous protein production in living animals in real-time, we use CRISPR-Cas9 genome-edited animals that have a PQR inserted into the gene encoding the Ribosomal protein L13a to track and measure protein expression dynamics in single cells *in vivo*. By imaging these animals *in vivo* and tracking protein expression over time in hundreds of single cells, we have found that protein synthesis events occur on timescales of tens of minutes but with large variability across single cells. Interestingly, we have found that cells that are closer together and from the same tissue have similar dynamics of protein production. Since we also created two different genome-edited animals that have different colors of the quantitative reporter, by crossing these two animals we can track protein synthesis from each parent-of-origin allele simply by using color. We have found that nearly all cells express both the mother's and father's copy of the *Rpl13a* gene, but not always equally. Protein synthesis from both parent-of-origin alleles occur with similar dynamics of frequency and duration. Clusters of cells can have a bias for one parental allele which varies across tissues and can change over timescales of hours. Our results indicate that protein expression dynamics across cells and between the parent-of-origin alleles are stochastic, but initially determined by progenitor cells.

Disclosures: C. Lo: None. **B.E. Chen:** None.

Poster

436. Staining and Imaging Techniques

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 436.11/WW27

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: Swedish RC Grant 2013-3105 and 2014-6215

Swedish Brain Foundation

Swedish Foundation for Strategic Research Grant RIF14-0078

Science for Life Laboratory Grant

Title: Simultaneous mapping and quantitation of neurotransmitters directly in tissue sections using mass spectrometry imaging

Authors: *P. E. ANDREN[1], M. SHARIATGORJI[1], A. NILSSON[1], E. FRIDJONSDOTTIR[1], L. KATAN[1], J. SAVMARKER[1], P. SVENNINGSSON[2], L. ODELL[1]
[1]Uppsala Univ., Uppsala, Sweden; [2]Karolinska Institutet, Stockholm, Sweden

Abstract: We here introduce a novel approach to image a large number of different neurotransmitters, their precursors and metabolites simultaneously from brain tissue sections. Without modification, most neurotransmitters are not directly detectable by matrix-assisted laser desorption ionization (MALDI)-mass spectrometry imaging (MSI) due to their poor ionization efficiency and the overlapping signals of isobaric compounds. To address the limited ionization and desorption of neurotransmitters, we developed a method for their *in situ* chemical derivatization and charge-tagging. Once modified, the small molecule target compounds are readily ionized and detected, enabling their identification and the quantitative mapping of their distributions by molecular-specific MALDI-MSI. Using the derivatization approach we were able to map numerous neurotransmitter systems, such as dopamine and serotonin, their precursors and most of their metabolites directly in brain tissue sections. For example, the precursor to dopamine, L-DOPA, was imaged for the first time simultaneously with dopamine, and twelve known dopamine metabolites were imaged, as well as three different L-DOPA metabolites. To further demonstrate the potential of the MALDI-MSI methodology we applied the strategy to Parkinson's disease experimental models. In summary, our methodology enables *in situ* mapping of connectivity patterns within and between neurotransmitter systems. Quantitation of functional neurotransmitter balances may be a useful approach in studies of neurodegenerative disorders but also in drug development as a biomarker-based rationale for targeted modulation of neurotransmitter networks.

Disclosures: P.E. Andren: None. **M. Shariatgorji:** None. **A. Nilsson:** None. **E. Fridjonsdottir:** None. **L. Katan:** None. **J. Savmarker:** None. **P. svenningsson:** None. **L. Odell:** None.

Poster

436. Staining and Imaging Techniques

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 436.12/WW28

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH grant R01-MH103160

NIH grant R01-NS076462

BRAIN Initiative award R24-MH109081

Title: Engineered hemodynamic imaging of neuropeptides and proteases in the brain

Authors: *M. DESAI[1], A. D. SLUSARCZYK[1], M. D. BARCH[1], A. D. CHAPIN, 02139[1], A. JASANOFF[2]
[2]Biol. Engin., [1]MIT, Cambridge, MA

Abstract: *In vivo* imaging of specific molecular processes across whole brains could lead to earlier diagnosis of neurological diseases, more effective drug evaluation, and advances in basic neurobiology, but molecular imaging techniques are constrained by limitations of the available probes. Magnetic resonance imaging (MRI) offers deeper tissue penetration than optical modalities and higher spatial resolution than nuclear imaging techniques, but molecular imaging agents for MRI can only be applied at levels to high for measurement of many species in the brain. Recently we demonstrated a new and highly sensitive approach for molecular-level imaging which entirely bypasses the need for conventional imaging contrast agents by perturbing the substantial reservoir of endogenous multimodal contrast afforded by the vasculature. We showed that imaging agents derived from calcitonin gene-related peptide (CGRP) can artificially induce vasodilation in rat brain and induce contrast changes, measured by optical imaging and MRI, at nanomolar concentrations in in the brain. Here we demonstrate that such agents can be used as building blocks for switchable analyte-dependent and genetically encoded reporters suitable for molecular imaging of proteases and neuropeptides at physiologically relevant concentrations. These tools offer new capability for monitoring dynamic aspects of brain biology, with potential applications in both science and medicine.

Disclosures: M. Desai: None. **A.D. Slusarczyk:** None. **M.D. Barch:** None. **A.D. Chapin:** None. **A. Jasanoff:** None.

Poster

436. Staining and Imaging Techniques

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 436.13/WW29

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIHDP2MH019427

Title: An axon-targeted GCaMP sensor for *In vivo* imaging of distal and local axons in mammalian cortex

Authors: *G. J. BROUSSARD, JR[1], Y. LIANG[2], M. FRIDMAN[3], E. K. UNGER[1], G. MENG[2], L. T. PETREANU[3], N. JI[2], L. TIAN[1]
[1]Univ. of California At Davis, Davis, CA; [2]Janelia Res. Campus, HHMI, Ashburn, VA; [3]Champalimaud Neurosci. Programmme, Lisbon, Portugal

Abstract: Understanding brain function demands linking neural activity from identified cell types in different brain areas with the animal's behavior. Optical recordings from distant axons terminated in the imaging area provide direct interrogation of output signals carried by specific afferents projected from distant neuronal populations. Untargeted Genetically-encoded- Ca^{2+}-indicators (GECIs) diffuse poorly to small presynaptic compartment in distal axons, resulting in dim, noisy signals. To facilitate optical recording in axons, we developed preSyn-GCaMP6, an axonally-targeted GECI employing a genetic strategy for active targeting to distal and local axons. We characterized the photochemical properties, targeting efficiency and sensitivity of preSyn-GCaMP6s in vitro and in vivo in mammalian neurons. PreSyn-GCaMP6s showed about 6-fold increased enrichment, 2-fold enhanced signal to noise ratio (SNR) and increased density of active boutons in thalamic axons in response to oriented gratings in mouse primary visual cortex (V1), compared to cytosolic GCaMP6s. The increased probe concentration resulted in larger baseline fluorescence in distal axons, facilitating recordings in vivo and image registration. In addition, we were able to image boutons at a depth of 600μm, approximately 50% deeper than anything previously reported with cytosolic GCaMP6s. We then utilize preSyn-GCaMP6s to record layer specific local circuit outputs while excluding postsynaptic signals. Using optical recording with preSyn-GCaMP6s, we discovered a previously unknown difference in tuning responses across cortical layers in axons projected from cells of layer 4 in V1.

Disclosures: G.J. Broussard: None. **Y. Liang:** None. **M. Fridman:** None. **E.K. Unger:** None. **G. Meng:** None. **L.T. Petreanu:** None. **N. Ji:** None. **L. Tian:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.01/WW30

Topic: I.04. Physiological Methods

Support: NIH Director's New Innovator 1DP2NS087949 and PECASE

NIH Grant SPARC 1OT2OD023848-01

NIH/National Institute on Aging Grant 1R01AG047664

NIH Grant BRAIN 1U01NS090577

Hereditary Disease Foundation

Friedreich's Ataxia Research Alliance (FARA) and FARA Australasia

Title: An AAV toolbox for enhanced transduction efficiency with regional and/or cell-type specificity in the CNS and PNS after systemic delivery

Authors: *K. CHAN, S. R. KUMAR, M. J. JANG, Y. LUO, R. C. HURT, N. C. FLYTZANIS, N. GOEDEN, B. E. DEVERMAN, V. GRADINARU
Biol. and Biol. Engin., Caltech, Pasadena, CA

Abstract: Recombinant adeno-associated viruses (rAAVs) are extensively used for *in vivo* gene transfer. Capsids and rAAV genomes that enable widespread and efficient gene expression following noninvasive administration with potential for expression that is restricted to target elements (cell types and/or defined nuclei) are needed. We therefore applied the capsid selection method CREATE (Cre-recombination-based AAV targeted evolution; Deverman et al., Nat. Biotechnol., 2016) to evolve and select for novel capsids that more efficiently or more selectively transduce specific cell populations. Here we report several novel capsids with neural tropisms. First, AAV-PHP.eB, an enhanced version of the previously reported AAV-PHP.B vector, is able to transduce 69% of $DAPI^+$ cortical cells after intravenous delivery of 1 x 10^{11} vg per adult mouse. Second, AAV-PHP.S, transduces neurons throughout the PNS. At a dose of 1 x 10^{12} vg per adult mouse, intravenous administration of AAV-PHP.S transduces 82% of $PGP95^+$ dorsal root ganglion neurons. In addition, because of the efficiency of AAV-PHP.S following systemic delivery, it is able to efficiently transduce difficult-to-access sensory neurons, such as those in the cardiac ganglia, or peripheral sensory networks that are widespread, such as the enteric nervous system. Third, we have identified an AAV-PHP.B variant, AAV-PHP.N, which shows strong selectivity for CNS neurons, and reduced liver transduction. To achieve targeted gene expression in nontransgenic animals, we are also presenting on the use of rAAV vectors with both cell type-specific promoters and with miRNA binding sites in the 3' UTR to reduce expression in specific neural cell types. By pairing previously described cell type specific promoters such as serotonergic (FEV), human synapsin 1 (hSyn1), interneuron (Dlx5/6), tyrosine hydroxylase (TH) and Purkinje Cell (PCP), with the AAV-PHP vectors, we can achieve both high efficiency and specificity of gene expression in the CNS and PNS. Finally, we have developed a two-component viral vector system to control the density of labeling when systemically delivering genes with AAV-PHP viruses. We demonstrate the utility of such a system by separately encoding spectrally distinct fluorescent proteins under various gene regulatory elements for single-cell morphology studies. Collectively, the versatility and efficiency of these viral capsids and viral vectors provides an expansion to the current AAV toolbox with potential for efficient and versatile gene manipulation studies.

Disclosures: K. Chan: None. **S.R. Kumar:** None. **M.J. Jang:** None. **Y. Luo:** None. **R.C. Hurt:** None. **N.C. Flytzanis:** None. **N. Goeden:** None. **B.E. Deverman:** None. **V. Gradinaru:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.02/WW31

Topic: I.04. Physiological Methods

Support: NIH Director's New Innovator IDP20D017782-01

PECASE

NIH/NIA 1R01AG047664-01

NIH BRAIN 1U01NS090577

Heritage Medical Research Institute

Pew Charitable Trust

Beckman Institute

Title: Deep brain optical imaging reveals motivational salience encoding by dorsal raphe dopamine neurons

Authors: *J. CHO[1], J. E. ROBINSON[2], J. B. TREWEEK[2], D. A. WAGENAAR[2], V. GRADINARU[2]
[1]Computation and Neural Systems, [2]Biol. and Biol. Engin., Caltech, Pasadena, CA

Abstract: Although reward and punishment have opposite valence and promote approach and avoidance respectively, both stimuli can enhance arousal and capture attention to guide appropriate behavioral responses. We have previously demonstrated that dopamine (DA) neurons in the dorsal raphe nucleus (DRN), which project densely to the extended amygdala, are wake-active over sleep states, promote arousal, and show robust activation to salient stimuli, irrespective of their hedonic valence [1]. We further examined whether DRN-DA neurons encode motivational salience [2] (n = 6 TH-Cre mice) with fiber photometry [3]. GCaMP6f-expressing mice were subjected to fear memory acquisition and extinction, where sensory cues of originally neutral context gain and lose motivational salience. Before conditioning, DRN-DA neurons showed small activation to the novel sensory cues (conditioned stimuli, CS; house-light and 65 dB 5 kHz tone) and no significant change across repetitive exposures (Pearson's $r = -0.12$, $p = 0.75$). Throughout learning and repeated pairings (10 trials, random ITIs within [70, 110] seconds) of CS and electric footshock (unconditioned stimuli, US; 0.6 mA for 1 sec), DRN-DA neurons gradually developed phasic response to the CS (Pearson's $r = 0.78$, $p < 0.01$). As previously shown [1], US induced robust activation of DRN-DA neurons, which however

decreased across repeated exposures (Pearson's $r = -0.90$, $p < 0.001$). In contrast to the learning phase, evoked response to the CS diminished across extinction trials, where sensory cues lost motivational salience (Pearson's $r = -0.83$, $p < 0.0001$). These findings suggest that phasic activation of DRN-DA groups encodes motivational salience and that they are modulated by expectations at the population level. To discriminate between projection-specific functions at the single-cell level, we built a two-photon microscope for deep brain imaging of projection-identified DRN-DA cell bodies during appetitive and aversive conditioning tasks. Taken together, these data deepen our understanding of the functional properties of DRN-DA neurons.

Reference:

[1] Cho JR, Treweek JB, Robinson JE, Xiao C, Bremner LR, Greenbaum A, Gradinaru V. (2017) Dorsal raphe dopamine neurons modulate arousal and promote wakefulness by salient stimuli. Neuron *in press*.

[2] Bromberg-Martin ES, Matsumoto M, Hikosaka O. (2010) Dopamine in motivational control: rewarding, aversive, and alerting. Neuron *68*, 815-834.

[3] Lerner TN, Shilyansky C, Davidson TJ, Evans KE, Beier KT, Zalocusky KA, Crow AK, Malenka RC, Luo L, Tomer R, Deisseroth K. (2015) Intact-brain analyses reveal distinct information carried by SNc dopamine subcircuits. Cell *162*, 635-647.

Disclosures: J. Cho: None. **J.E. Robinson:** None. **J.B. Treweek:** None. **D.A. Wagenaar:** None. **V. Gradinaru:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.03/WW32

Topic: I.04. Physiological Methods

Support: NIH Director's New Innovator Award/PECASE IDP20D017782-01

NIMH under Ruth L. Kirschstein National Research Service Award F31MH102913

NIGMS under Ruth L. Kirschstein National Research Service Award F32GM116319

V.G. is a Heritage Principal Investigator supported by the Heritage Medical Research Institute

the Beckman Institute for the Resource Center on CLARITY, Optogenetics, and Vector Engineering for technology development and broad dissemination (http://www.beckmaninstitute.caltech.edu/clover.shtml)

NIMH Grant R21MH103824

Title: Machine learning to predict expression, membrane localization, and functional properties of diverse Channelrhodopsin variants

Authors: *C. BEDBROOK[1], K. K. YANG[2], A. J. RICE[2], X. DING[1], F. H. ARNOLD[2], V. GRADINARU[1]
[1]Biol. and Biol. Engin., [2]Chem. and Chem. Engin., CALTECH, Pasadena, CA

Abstract: To facilitate neuronal circuit interrogation, we aim to advance the repertoire of current optogenetic tools, focusing on diversifying spectral properties, kinetics, and increasing current strength of the light-gated channel, Channelrhodopsin (ChR). To engineer integral membrane ChRs we must introduce mutations that improve desired functional properties while retaining expression and membrane localization. This is a major challenge because the sequence and structural determinants of ChR expression and localization are highly constrained and largely unknown. To address this challenge we trained data-driven models to predict ChR sequences that will express, localize, and function. To train these models we used a data set of expression, localization and functional properties of >200 ChR sequences of our own design [1] combined with data from previously published ChR variants [2,3]. For our designs we used structure-guided SCHEMA recombination [4] to create a large ($2x10^3$) set of functionally diverse chimeras from three sequence-diverse ChRs (CsChrimsonR, C1C2, and CheRiff). From this recombination library we chose 218 ChR chimeras and assayed them for expression and plasma membrane localization in mammalian cells. The majority of the ChR chimeras express and a significant fraction also localize to the membrane. The majority of these localizing chimeras are also functional light-gated channels. Many chimeras have stronger light-activated inward currents than the three parents, and some have unique off-kinetics and spectral properties relative to the parents. Combining these data with the published data on different ChR variants we were able to build models that reliably predict ChR properties. We use these models to identify sequence diverse ChRs capable of efficient localization and function. We applied these models to convert naturally occurring ChR incapable of mammalian localization into ones that localizes well and have novel functional properties. We can interpret these models to elucidate sequence and structure elements important for ChR expression, membrane localization, and functional properties to inform further engineering efforts.
1. Bedbrook CN, et al. Structure-guided SCHEMA recombination generates diverse chimeric channelrhodopsins. PNAS. 2017.
2. Mattis J, et al. Independent optical excitation of distinct neural populations. Nature methods. 2011
3. Klapoetke NC, et al. Principles for applying optogenetic tools derived from direct comparative analysis of microbial opsins. Nature methods. 2014.
4. Voigt CA, et al. Protein building blocks preserved by recombination. Nature structural biology. 2002.

Disclosures: C. Bedbrook: None. **K.K. Yang:** None. **A.J. Rice:** None. **X. Ding:** None. **F.H. Arnold:** None. **V. Gradinaru:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.04/WW33

Topic: I.04. Physiological Methods

Support: NIH Director's New Innovator Award/PECASE IDP20D017782-01

NIMH under Ruth L. Kirschstein National Research Service Award F31MH102913

V.G. is a Heritage Principal Investigator supported by the Heritage Medical Research Institute

the Beckman Institute for the Resource Center on CLARITY, Optogenetics, and Vector Engineering for technology development and broad dissemination (http://www.beckmaninstitute.caltech.edu/clover.shtml)

Title: Engineering prokaryotic repressors for optical sensing of neurotransmitters

Authors: *X. DING, C. N. BEDBROOK, N. HUTCHINS, V. GRADINARU
Biol. and Biol. Engin., Caltech, Pasadena, CA

Abstract: As chemical transmission plays a critical role in neural communication, there is a growing need for tools that can report on the dynamics of neurotransmitters *in vivo*. Here we explored the use of prokaryotic transcriptional repressors, specifically TetR family repressors, for sensing melatonin and N-acetylserotonin (NAS). The TetR family of repressors is a highly diverse family of genetic regulators that share the same general fold but sense a large variety of bio-active small molecules. We first screened a number of these natural TetR proteins and identified one TetR family repressor that showed promiscuous sensitivity to melatonin in millimolar (mM) concentrations. Using directed evolution, we improved the repressor's sensitivity and specificity to sense melatonin in micromolar (μM) concentrations, which is within the physiological levels in mammalian pineal glands. In each round of evolution, libraries comprising 1000 – 20,000 variants generated with error-prone PCR or site saturation mutagenesis were transformed into *E.coli* and screened with a high-throughput fluorescence-based assay. Hits with greatest improvement in both sensitivity and specificity were selected, validated, and further evolved. In addition to melatonin-responsive repressors, we also identified NAS-responsive repressors, that could enable multiplexed monitoring of both melatonin and NAS. Interestingly, some repressor variants show reversed ON-OFF activity, which leads to the potential of engineering reverse melatonin-controlled transactivators. Both the repressors and the directed evolution method presented here provide a promising platform for development of genetically encoded neurotransmitter sensors and targeted expression of other neural modulators.

Disclosures: **X. Ding:** None. **C.N. Bedbrook:** None. **N. Hutchins:** None. **V. Gradinaru:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.05/WW34

Topic: I.04. Physiological Methods

Support: John P. Stock Fellowship

Title: Elucidating the circuit architecture and function of NAc inputs to the VTA

Authors: H. YANG, J. W. DE JONG, Y. TAK, J. R. PECK, H. BATEUP, *S. LAMMEL
Mol. and Cell Biol., UC Berkeley, Berkeley, CA

Abstract: The nucleus accumbens (NAc) is a crucial brain structure implicated in reward processing and neuropsychiatric disorders. In contrast to ventral tegmental area (VTA) dopamine (DA) neurons projecting to NAc, which have been intensively studied over the last decades, much less is known about the circuit architecture and functional role of NAc inputs to the VTA. Recently, anatomical tracing and electrophysiological studies generated considerable debate regarding the synaptic connectivity of inhibitory NAc inputs with DA and non-DA VTA neurons. Importantly, a major limitation of previous work is that the both NAc and VTA have been considered to be largely homogeneous brain structures. Using a multidisciplinary approach combining retrograde tracing, *ex vivo* electrophysiology and optogenetics, we investigated whether distinct subdivisions of the NAc (medial shell [NAcMed] versus lateral shell [NAcLat]) target different DA and non-DA subpopulations in the VTA. Our data demonstrate that D1-expressing medium spiny neurons in NAcMed and NAcLat target different VTA subregions. NAcMed terminals are located preferentially in the medial VTA, while NAcLat terminals can be found in the lateral VTA and substantia nigra. Next, we elucidated the circuit architecture of NAcMed and NAcLat inputs to the VTA. We found that NAcMed inputs predominantly exert direct inhibitory, GABA-A receptor-dependent control over VTA DA neurons projecting to NAcMed. In contrast, NAcLat inputs preferentially inhibit VTA GABA neurons, which presumably results in a disinhibition of DA neurons projecting to NAcLat, as optogenetic stimulation of NAcLat terminals in the VTA increases neural firing of DA neurons projecting to NAcLat. Consistent with a potential mechanism for disinhibition is the finding that *in vivo* optogenetic stimulation of NAcLat terminals in the VTA could strongly drive instrumental behavior as well as real-time place preference. Collectively, these observations strongly suggest that the mesolimbic DA system is comprised of discrete circuits, which may serve distinct biological functions in the regulation of motivated behaviors.

Disclosures: **H. Yang:** None. **J.W. de Jong:** None. **Y. Tak:** None. **J.R. Peck:** None. **H. Bateup:** None. **S. Lammel:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.06/WW35

Topic: I.04. Physiological Methods

Support: HHWF

NRSA

NIH

DARPA

Title: Causal link between neocortical single-cell ensemble activity and specific behaviors

Authors: ***J. H. JENNINGS**[1], C. K. KIM[1], J. MARSHEL[1], M. RAFFIEE[1], L. YE[1], S. QUIRIN[1], S. PAK[1], C. RAMAKRISHNAN[1], K. DEISSEROTH[1,2,3]
[1]Bioengineering, Stanford Univ., Palo Alto, CA; [2]Psychiatry & Behavioral Sci., Stanford Univ., Stanford, CA; [3]Howard Hughes Med. Inst., Stanford, CA

Abstract: Neurons in orbitofrontal cortex (OFC) individually respond differentially to distinct rewarding experiences such as caloric- or social-related stimuli, but it is unknown if these activity-specific ensembles play causal roles in discrete behaviors, or in circuit dynamics. To dissect individual contributions of activity-specific OFC neuronal populations to behavior and circuit physiology, we coupled genetically encoded Ca^{2+} imaging with engineered microbial opsin-based control, to optically both monitor and manipulate activity of OFC neurons at the single cell level with two-photon microscopy. Employing this all-optical approach in head-fixed mice during caloric consumption or social interaction, we identified a spatially-biased clustered population within OFC (caloric-encoding cells mean distance=97.66μm ±3.09 SEM, random cells mean distance=110.40μm ±2.30 SEM, P=0.005) that selectively responded to caloric rewards and displayed minimal overlap with social-specific cells ($F_{2,30}$=20.79, P<0.0001, mean=25% caloric-only, 11% social-only, and 6% overlapping (both) cells, n=11 mice). Next, two-photon optogenetic stimulation of individual caloric-responsive neurons was found to play a causal role in behavior, significantly increasing consumption compared to non-stim sessions ($F_{2,30}$=145.7, P<0.0001, mean=20 stim cells, n=6 mice) and to control mice lacking an opsin ($F_{2,30}$=127.9, P<0.0001, mean=20 stim cells, n=6 mice per group); conversely, single cell activation of social-specific neurons did not alter caloric intake ($F_{2,30}$=0.444, P=0.9918, mean=20 stim cells, n=6 mice). We next sought to determine local network activity dynamics arising from

specific single cell-resolution stimulation. The number of neurons that were optogenetic stimulus-excited but not directly-targeted increased monotonically with the number of successfully-targeted cells (Spearman's rho=0.82, P=0.02); surprisingly, this relationship was not seen for neurons that were optogenetic stimulus-inhibited but not directly-targeted (Spearman's rho=-0.14, P=0.78) (*n*=10 light targets per mouse, *n*=9 mice). Of interest, the excited but non-directly-targeted cells exhibited detectable spatial clustering within OFC (excited cells mean distance=80.34 ±6.57 SEM, random cells mean distance=113.53 ±5.26 SEM, P=0.008). Taken together, using all-optical methods for cellular-resolution behaviorally-relevant ensemble control and imaging, we have identified an activity-specific ensemble that is functionally and spatially distinct within OFC that locally modulates other spatially-distinct clusters and selectively controls the behavior it encodes.

Disclosures: J.H. Jennings: None. **C.K. Kim:** None. **J. Marshel:** None. **M. Raffiee:** None. **L. Ye:** None. **S. Quirin:** None. **S. Pak:** None. **C. Ramakrishnan:** None. **K. Deisseroth:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.07/WW36

Topic: I.04. Physiological Methods

Support: 1F32MH110144

R01DA03537701

Title: Cell-type specific reward dynamics of habenular neurons

Authors: *E. L. SYLWESTRAK[1], S. VESUNA[2], A. CROW[1], C. RAMAKRISHNAN[2], K. DEISSEROTH[3]
[1]Bioengineering, [2]Stanford Univ., Stanford, CA; [3]Bioengin & Psych, Stanford Univ. Dept. of Psychology, Stanford, CA

Abstract: The processing of appetitive and aversive stimuli is essential to guide motivated behavior and is represented in many structures across the mammalian brain. The habenular complex is comprised of a transcriptionally diverse group of cells in the dorsal thalamus, divided into two anatomically distinct regions. The medial subdivision receives input from the limbic system and send projections to the midbrain, and has been shown to play a role in behaviors related to processing of aversive and appetitive stimuli, including stress, nicotine withdrawal, and voluntary exercise. This region is smaller and less studied than its lateral counterpart, but it displays a strong degree of transcriptional heterogeneity, with cells expressing a wide variety of neuromodulatory receptors, neurotransmitters, and neuropeptides. Using multiplexed

transcriptional analysis, viral projection mapping and tissue clearing, we analyzed the distribution of Th-, ChAT- and Substance P- expressing neurons in the habenula and their axonal termination patterns in the midbrain. Using Cre-mediated viral delivery in subpopulations of medial habenula neurons and fiber-based calcium imaging during an operant task, we find that habenular subpopulations exhibit cell-type specific reward dynamics: tyrosine hydroxylase expressing neurons only respond to reward-predictive cues (n=4 mice, p=0.02), cholinergic neurons are not significantly modulated by reward (n=3 mice, p=.33), and tackykinin neurons respond to both reward and reward prediction error (n=6 mice, p=0.01), but with distinct temporal dynamics. These data suggest that medial habenula neurons encode multiple aspects of reward responses and could serve as a mechanism to modulate motivated behavior in different emotional states.

Disclosures: E.L. Sylwestrak: None. **S. Vesuna:** None. **A. Crow:** None. **C. Ramakrishnan:** None. **K. Deisseroth:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.08/WW37

Topic: I.04. Physiological Methods

Support: NSF IGERT

HHMI

NSF

Title: All-optical closed-loop feedback control of targeted neuronal populations in awake animals

Authors: *N. YOUNG, C. K. KIM, M. INOUE, Y. S. KIM, K. DEISSEROTH
Stanford Univ., Stanford, CA

Abstract: Stimulation of targeted neural cells and tissues can be achieved with varying degrees of temporal precision and cell type specificity. Most stimulus strategies used today are “open-loop” in that they ignore ongoing activity and other features of the momentary brain state. For many applications, however, it is desirable to make real-time adjustments to stimulus intensity and waveform based on ongoing activity in the same or other brain areas[1], though this has proven difficult to achieve with electrical methods due to large stimulation artifacts and a lack of cellular specificity. Here we present an all-optical setup for applying stimulation to defined subsets of neurons in closed-loop, leveraging the advantages of genetically-targeted population recording/control. We co-expressed spectrally-separated opsin and fluorescent Ca^{2+} indicator in

genetically-defined subpopulations of neurons in behaving mice. Ca^{2+} responses (serving as a proxy for cellular activity) were recorded using the Frame-Projected Independent-Fiber Photometry (FIP) method described previously[2]. Opsin excitation was achieved by laser through the same optics used for activity recording. This system exhibited a stimulation artifact below 3% of total signal. Surprisingly, we found that simple PID control was sufficient to bring activity within 20% dF/F of a predetermined setpoint. These results demonstrate the utility of approximating population neural activity (as a single time-varying scalar) in behaving animals with a linear plant model.

References:
1. Grosenick L, Marshel JH, Deisseroth K. (2015) Neuron.
2. Kim CK, et al. (2016) Nature Methods.

Disclosures: **N. Young:** None. **C.K. Kim:** None. **M. Inoue:** None. **Y.S. Kim:** None. **K. Deisseroth:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.09/WW38

Topic: I.04. Physiological Methods

Support: NIH Grant F31DA041795 to C.K.K.

Helen Hay Whitney Foundation to J.H.J.

NIMH and DARPA to K.D.

Title: Molecular and circuit-dynamical identification of top-down neural mechanisms for restraint of reward-seeking

Authors: *C. K. KIM, L. YE, J. H. JENNINGS, N. PICHAMOORTHY, D. D. TANG, C. RAMAKRISHNAN, A.-C. WANG, K. DEISSEROTH
Stanford Univ., Stanford, CA

Abstract: Reward-seeking behavior is fundamental to survival, but suppressed acquisition of even highly-valued rewards can be essential as well; such suppression is adaptive in diverse naturally-occurring situations and is thought to be disrupted in individuals suffering from addiction and other major neuropsychiatric diseases. In humans and rodents, the medial prefrontal cortex (mPFC) has been implicated in suppressing both natural and compulsive reward-seeking; however, despite vital significance in health and disease, the neural circuit pathways through which mPFC regulates reward-seeking remain to be found. Here we show that a specific subset of superficial mPFC projections to a subfield of nucleus accumbens (NAc)

neurons naturally encodes the decision to initiate or suppress reward-seeking when faced with harmful consequences (81.05±1.30% prediction accuracy, bootstrapping with 1,000 shuffles, $p<0.05$, $n=5$ mice). Surprisingly, mPFC projections to the ventral tegmental area- source of the best understood and most potent reward circuitry in the vertebrate brain- did not encode (and could not causally elicit) suppression of reward-seeking; moreover, while optogenetic stimulation of mPFC→NAc neurons as a population could elicit aversion of a neutral environment (Paired t-test, $t_7=2.67$, $p=0.032$, $n=8$ mice), suppression of reward-seeking by this mPFC→NAc population was not found, strikingly even when reward value was mitigated by association with a harmful stimulus (Tukey-Kramer test on 1-way ANOVA, $p=0.69$, $n=8$ mice). Only a highly-resolved subpopulation of top-down projecting neurons, identified by 2-photon Ca^{2+} imaging and novel activity-dependent labeling strategies to recruit the relevant subset of neurons, was capable of suppressing reward-seeking (2-way ANOVA interaction, $F_{1,10}=6.79$, $p=0.026$, $n=6$ mice). This natural activity-resolved mPFC projection to NAc displayed unique molecular-genetic and microcircuit-level features concordant with a broad role in the conserved behavior of reward-seeking regulation, providing cellular and anatomical identifiers of behavioral and possible therapeutic significance.

Disclosures: **C.K. Kim:** None. **L. Ye:** None. **J.H. Jennings:** None. **N. Pichamoorthy:** None. **D.D. Tang:** None. **C. Ramakrishnan:** None. **A. Wang:** None. **K. Deisseroth:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.10/WW39

Topic: I.04. Physiological Methods

Support: NIH Grant 5U01NS099573

Title: Engineering of near-infrared genetically encoded voltage indicators

Authors: M. MONAKHOV[1], M. MATLASHOV[2], D. SHCHERBAKOVA[2], A. BOILLAT[3], C. SONG[3], S. ANTIC[1], V. VERKHUSHA[2], *T. KNOPFEL[3,4]
[1]Inst. for Systems Genomics, Stem Cell Inst., UConn Hlth., Farmington, CT; [2]Albert Einstein Col. of Med., New York, NY; [4]Ctr. for Neurotechnology, [3]Imperial Col. London, London, United Kingdom

Abstract: Simultaneous optical modulation and readout of neuronal circuit activities is a promising neurotechnology, which could help us decipher how the brain's electrical signals relate to perceptual, cognitive, emotional and motor functions. During recent years, the use of genetically encoded (optogenetic) actuators such as channelrhodopsin, became overwhelmingly successful. On the other hand, genetically encoded voltage indicators (GEVIs) have not yet been

satisfactorily optimized and their combination with optogenetic modulation has been difficult to achieve in practice. One major obstacle is the overlap of the spectral bands of light used to activate opsin-based actuators and at the same time excite and image available GEVIs. We propose to use novel bacteriophytochrome-based fluorescent proteins (FPs) to generate a new class of GEVIs that are excited and emit fluorescence in the near-infrared (NIR) spectrum (e.g. 720 nm). For this, we have engineered and characterized a set of spectrally diverse monomeric NIR FPs, termed miRFPs, and produced an effective FRET pair consisting of miRFP670 and miRFP720 proteins. Using this FRET pair we then designed several ratiometric FRET-based NIR GEVI constructs. We have also engineered several intensiometric NIR GEVI variants based on single NIR FPs, either miRFP703 or miRFP720. Both types of the NIR GEVI constructs demonstrated good plasma membrane localization and exhibited up to 3% ΔF/F in a response to a 100 mV depolarizing step in membrane voltage in patch-clamped HEK293 cells. To further optimize these NIR GEVI constructs, we apply an all-optical screening platform consisting of imaging hardware and mammalian cells stably expressing optogenetic actuators.

Disclosures: **M. Monakhov:** None. **M. Matlashov:** None. **D. Shcherbakova:** None. **A. Boillat:** None. **C. Song:** None. **S. Antic:** None. **V. Verkhusha:** None. **T. Knopfel:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.11/WW40

Topic: I.04. Physiological Methods

Support: John P. Stock Fellowship

Brain & Behavior Research Foundation

Brain Research Foundation

Title: Neural circuit mechanisms underlying drug-induced changes in motivated behaviors

Authors: *J. W. DE JONG[1], S. A. AFJEI[1], J. R. PECK[1], V. HAN[1], C. K. KIM[2], K. DEISSEROTH[3], S. LAMMEL[1]
[1]Mol. and Cell Biol., UC Berkeley, Berkeley, CA; [2]Stanford Univ., Stanford, CA; [3]Bioengin & Psych, Stanford Univ. Dept. of Psychology, Stanford, CA

Abstract: The mesocorticolimbic dopamine (DA) system, which is comprised of DA neurons in the ventral tegmental area (VTA) and their projections to different forebrain regions, has been fundamental in the formulation of most models of drug use, abuse, and addiction. Recently, we demonstrated that different afferent inputs to the VTA mediate reward- and aversion-related behaviors in profoundly different ways. Because drugs of abuse (e.g., cocaine, morphine) modify

the function of excitatory inputs to the VTA, we hypothesized that drug-evoked synaptic adaptations in specific VTA afferent pathways may underlie some of the maladaptive behaviors of individuals with substance use disorder (SUD). The lateral hypothalamus (LH) represents a major afferent pathway of the VTA, which has been previously implicated in both positive and negative motivational states. Using a multidisciplinary approach combing *in vivo* optogenetics, fiber photometry, synaptic electrophysiology, rabies virus-based tracing and *in situ hybridization*, we studied a) function, b) circuit architecture and c) drug-evoked synaptic plasticity of the LH→VTA pathway. We first established a critical role of the glutamatergic component of the LH→VTA pathway for mediating aversion-related behaviors by performing optogenetic stimulation and silencing experiments as well as fiber photometry in freely moving mice. Next, we elucidated the complete circuit architecture of the LH→VTA pathway by combining anatomical and functional neural circuit mapping. We found that, qualitatively, LH neurons target all VTA DA and non-DA subpopulations. Quantitatively, however, we discovered a highly-biased input scheme, which reveals that DA neurons projecting to nucleus accumbens (NAc) medial shell represent a dominant downstream effector of glutamatergic LH neurons. Remarkably, 24 hours after a single injection of cocaine (15 mg/kg, intraperitoneal), the synaptic strength of excitatory LH inputs onto VTA DA neurons projecting to NAc medial shell was significantly reduced compared to saline-treated animals. In contrast, a single injection of cocaine had no effect on the synaptic strength of excitatory LH inputs onto VTA DA neurons projecting to NAc lateral shell. Taken together, our results suggest that cocaine exposure induces synaptic depression in a brain pathway that encodes aversion-related behaviors, which may explain some of the maladaptive behaviors of individuals with SUD in which they continue drug-seeking/drug-taking behavior in the face of negative consequences.

Disclosures: **J.W. De Jong:** None. **S.A. Afjei:** None. **J.R. Peck:** None. **V. Han:** None. **C.K. Kim:** None. **K. Deisseroth:** None. **S. Lammel:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.12/WW41

Topic: I.04. Physiological Methods

Support: A.P. Giannini Foundation Postdoctoral Fellowship

Title: Amygdalonigral salience signals shape action-outcome associations

Authors: *E. E. STEINBERG[1], F. GORE[1], B. D. HEIFETS[1], K. T. BEIER[1], C. FÖLDY[2], T. N. LERNER[3], M. D. TAYLOR[1], L. LUO[1], K. DEISSEROTH[1], R. C. MALENKA[1]
[1]Stanford Univ., Palo Alto, CA; [2]Brain Res. Inst., Univ. of Zurich, Zurich, Switzerland; [3]Northwestern Univ., Chicago, IL

Abstract: The central amygdala (CeA) is known to orchestrate learned fear responses in many species. In addition to its well-described role in fear, the CeA also influences other cognitive processes including reward learning. However, the specific neural circuits that mediate CeA-dependent reward processes and their relationship to fear-promoting circuits remain poorly defined. Midbrain dopamine neurons perform essential computations that are required for many aspects of reward learning. Accordingly, we hypothesized that the robust projection from the CeA to the substantia nigra pars lateralis - a heterogeneous region containing dopamine, GABA and glutamate neurons - was well-positioned to link CeA signals with downstream effectors of reward learning. Consistent with our hypothesis, gain-of-function experiments revealed that optogenetic activation of amygdalonigral projections could potently drive instrumental (action-outcome), though not Pavlovian (cue-outcome), associations, pointing to a selective role for amygdalonigral neurons in reinforcement learning. However, single-cell gene expression analyses demonstrated that amygdalonigral neurons overlap with genetically defined cell types previously implicated in fear learning. Using fiber photometry to monitor population activity during natural behavior, we found that amygdalonigral neurons were activated by both appetitive and aversive stimuli during action-outcome and cue-outcome learning, indicating that as a population, amygdalonigral neurons signal salience as opposed to valence. Ongoing experiments seek to establish synaptic and circuit mechanisms that enable amygdalonigral signals to influence future behavior. Taken together, our data suggest that amygdalonigral neurons encode a salience signal that engages strong motivational drives capable of profoundly shaping action selection, revealing a previously unappreciated mechanism for linking emotion, motivation and action that provides new insight into the complex neural interactions that endogenously regulate motivated behaviors.

Disclosures: E.E. Steinberg: None. **F. Gore:** None. **B.D. Heifets:** None. **K.T. Beier:** None. **C. Földy:** None. **T.N. Lerner:** None. **M.D. Taylor:** None. **L. Luo:** None. **K. Deisseroth:** None. **R.C. Malenka:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.13/WW42

Topic: I.04. Physiological Methods

Support: Helen Hay Whitney Foundation

NARSAD Young Investigator Award

Hertz fellowship

HHMI

NIMH

NIDA

DARPA

Title: Ancestral circuits for the coordinated modulation of brain state

Authors: *M. LOVETT-BARRON[1], A. ANDALMAN[1], W. E. ALLEN[1,2], S. VESUNA[1], I. KAUVAR[1,3], V. M. BURNS[1,4], K. DEISSEROTH[1,5,6]
[1]Bioengineering, [2]Neurosci. Program, [3]Electrical Engin., [4]Chem. and Systems Biol., [5]Psychiatry and Behavioral Sci., [6]Howard Hughes Med. Inst., Stanford Univ., Palo Alto, CA

Abstract: Internal brain states fluctuate during behavior, impacting perception, cognition, and action. Internal states are governed in part by neuromodulation, but the set of neuromodulatory cell types involved are not fully known. Here we screened for neuromodulatory neuron activity in larval zebrafish during a behavioral task that reports fluctuations in the internal state of alertness. We developed a method to classify cell types from brain-wide neural activity imaging through post hoc immunohistochemistry and volume registration, allowing us to record from neurons in 22 neuromodulatory nuclei during behavior. In addition to noradrenergic neurons in the locus coeruleus, we identified several unexpected cell types encoding alertness, including cocaine- and amphetamine-regulated transcript and cholinergic neurons in the tegmentum, as well as dopaminergic, serotonergic, and neuropeptide-Y-expressing cells in the hypothalamus. We also found somatostatin-expressing neurons in the hypothalamus correlated with reduced alertness. Alertness-encoding cell types were preferentially correlated with one another, and their cooperative activity permitted improved prediction of behavioral performance. We then used this dataset to guide targeted investigation of homologous neuromodulatory nuclei in mice. Deep brain in vivo calcium imaging revealed striking conservation of function between zebrafish and mice, across all seven identified neuromodulatory cell types tested. We found that optogenetic activation of any cell type correlated with alertness was sufficient to enhance alertness - detected by decreased reaction time. However, inactivation of any individual cell type did not change reaction time, suggesting partial redundancy in this behavioral context. Interestingly, we found that optogenetic inactivation of these same cell types in the absence of a motivated behavioral task could decrease physiological measures of alertness and behavioral indices of anxiety, suggesting that the overlap in cell type significance is dynamically conditional upon behavioral context and organismal need. These results reveal an evolutionarily conserved network of multiple neuromodulatory systems that cooperatively regulate a fundamental internal state.

Disclosures: M. Lovett-Barron: None. **A. Andalman:** None. **W.E. Allen:** None. **S. Vesuna:** None. **I. Kauvar:** None. **V.M. Burns:** None. **K. Deisseroth:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.14/WW43

Topic: I.04. Physiological Methods

Support: Helen Hay Whitney Foundation

NARSAD

AWS

NIMH

NSF

NIDA

DARPA REPAIR Program

Title: Habenular cell ensemble activity encoding adverse experience: Temporal tiling, brainwide response orchestration and behavioral state transitions

Authors: *A. S. ANDALMAN[1], V. M. BURNS[2], M. LOVETT-BARRON[3], M. BROXTON[4], B. POOLE[4], S. J. YANG[5], L. GROSENICK[6], T. N. LERNER[10], P. MOURRAIN[7], M. LEVOY[8], K. DEISSEROTH[9]
[1]Bioengineering, Neurosci. and CNC Program, [2]Chem. and Systems Biol., [3]Bioengineering, [4]Computer Sci., Stanford Univ., Palo Alto, CA; [5]Electrical Engin., [6]Bioengineering, Stanford Univ., Stanford, CA; [7]Psychiatry and Behavioral Sci., Stanford Univ., Palo Alto, CA; [8]Computer Sci., [9]Bioengineering & Psychlogy, Stanford Univ., Stanford, CA; [10]Physiol., Northwestern, Chicago, IL

Abstract: Prior experiences affect how future actions are evaluated and selected. This effect is central to reinforcement learning systems and is critical for generating adaptive behavior. Behavioral challenge assays use inescapable stressors to probe how the repeated failure of escape behaviors effect action selection; these assays cause animals to transition to a passive coping state in which minimal effort to escape is made. Several brain regions, including the raphe nucleus, habenula, and periaqueductal gray, are known to be important in regulating passive behavior, but it remains unclear where or how evidence from the ongoing behavioral challenge experience is accumulated to cause the transition from active to passive coping. Here, we develop a behavioral challenge protocol that induces passive coping in late-stage larval zebrafish and use this protocol to perform an unbiased brain-wide search for regions where neural activity

is correlated with the extent of behavioral challenge. We found that during behavioral challenge, individual habenular neurons exhibited staggered recruitment into an activated ensemble. This recruitment caused a steady rise in average neural activity that is confined to the habenula (35% Δf/f change by end of the 36 minute protocol; n=8 fish) and that could be bi-directionally modulated. Prior treatment with the fast-acting anti-depressant ketamine prevented this rise (2% Δf/f change; n=4), while repeat-exposure to behavioral challenge enhanced it (80% Δf/f change; n=12). The magnitude of this habenular signal was correlated with size of the behavior change ($r = -0.56$; $p = 0.003$). Outside the habenula, the raphe nucleus and the dorsal thalamus both exhibited significant reduction in activity which plateaued 22 minutes into the protocol (-19% and -20% Δf/f change, respectively; n=8). Optogenetic stimulation of the habenula combined with brain-wide imaging revealed that habenula activation was sufficient to reduce mobility in an aversive experience-dependent fashion, (-0.45 mm/s; n=8) as well as selectively reduce activity in the downstream raphe nucleus. Taken together, our results suggest that a unique pattern of staggered progressive population recruitment within the habenula encodes aversive experience and underlies behavioral pattern selection in the face of adverse conditions. The development of the larval zebrafish as a vertebrate system for studying active to passive behavioral state transitions creates new possibilities, both for genetic and pharmaceutical screens, and for understanding how the diverse brain areas involved in stress-coping interact-- both in normal and pathological states.

Disclosures: **A.S. Andalman:** None. **V.M. Burns:** None. **M. Lovett-Barron:** None. **M. Broxton:** None. **B. Poole:** None. **S.J. Yang:** None. **L. Grosenick:** None. **T.N. Lerner:** None. **P. Mourrain:** None. **M. Levoy:** None. **K. Deisseroth:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.15/WW44

Topic: I.04. Physiological Methods

Support: NIH-grant 5R01MH075957

HHMI

Title: A novel red-shifted excitatory channelrhodopsin with multiple properties enabling markedly improved integration of Ca^{2+} imaging with optogenetic control

Authors: *Y. KIM[1], M. INOUE[1], C. RAMAKRISHNAN[1], H. KATO[1], S. YOSHIZAWA[2], K. DEISSEROTH[1]
[1]Stanford Univ., Stanford, CA; [2]Univ. of Tokyo, Tokyo, Japan

Abstract: Abstract
A rapidly-evolving goal of modern neuroscience is to combine the power of optogenetic activity-control with the information on specific activity patterns that is accessible via genetically encoded fluorescent activity-sensors. This goal has been challenging to fully achieve, due in part to the overlapping spectra of actuators and sensors. Here, we report a novel excitatory channelrhodopsin (ChR) nicknamed here 'MO20' that was discovered through sequence analysis and biophysical characterization of rhodopsin-like proteins from over 600 microbial organisms. MO20 showed robust excitation by redshifted light in the range of $\lambda \sim 585$-650nm, suitable for compatibility with GFP-based activity sensors with minimal optical cross-talk. Peak photocurrent values under orange ($\lambda \sim 585$nm) and red ($\lambda \sim 650$nm) light were 4.5±0.6 and 1.0±0.16 nA, respectively; these values were significantly higher than for previously characterized red-shifted ChRs under the same illumination conditions when tested in parallel. Speed was also suitable; MO20 exhibited more than 80% spiking fidelity in trains at frequencies up to 40Hz (orange light); and action potentials could be readily induced in neurons with orange light with irradiance values down to 0.005mW/mm^2. In a neuronal culture imaging setting with the Ca^{2+} sensor GCaMP6m, MO20 showed 10-fold faster rise-and-decay Ca^{2+} signal kinetics and significantly higher fluorescent signal amplitude than the leading red-shifted ChRs. This striking difference likely relates to reduced spectral overlap between opsin and sensor, faster channel kinetics, larger photocurrent, and pH independence. These unprecedented biophysical properties define a tool for more powerful and precise probing of neuronal function via integration activity sensors and actuators.

Disclosures: Y. Kim: None. **M. Inoue:** None. **C. Ramakrishnan:** None. **H. Kato:** None. **S. Yoshizawa:** None. **K. Deisseroth:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.16/WW45

Topic: I.04. Physiological Methods

Support: Defense Advanced Research Projects Agency Neuro-FAST program

Title: Wide field-of-view three dimensional all-optical neurophysiology with millisecond-resolution *In vivo*

Authors: *J. H. MARSHEL, S. QUIRIN, E. L. SYLWESTRAK, A. CHIBUKHCHYAN, A. CROW, C. RAMAKRISHNAN, K. DEISSEROTH
Stanford Univ., Stanford, CA

Abstract: Of presumptive importance for neural coding and processing, cellular activity patterns are sparse in space and time within dense populations of neurons; neighbors often encode different information, have different inputs and postsynaptic partners, and are active at distinct points on the millisecond-timescale. To help identify and understand causal roles, on circuit dynamics and behavior, of precise activity patterns within functionally characterized subsets of neurons, we developed an all-optical microscopy system capable of optogenetic stimulation (e.g., using excitatory opsin $C1V1_{T/T}$) of dozens or more single neurons every 30 ms, addressing sequences of subpopulations with millisecond (~1-3 ms) precision in three dimensions while recording Ca^{2+} activity (GCaMP) of the same neurons and surrounding populations of at least hundreds of neurons in parallel. This novel system combines 3D two-photon imaging with 3D spatial light modulator (SLM) two-photon holographic stimulation, and among other features, improves power efficiency across the volume to quadruple the simultaneously addressable population of neurons (450x450μm x 100μm axial), when applied with standard pixel count SLMs (512x512) and high NA objectives (25x 1.05 NA). We further demonstrate how to at-least double the ensemble excitation pattern-refresh rate of SLM systems, which until now have been theoretically limited to 10-100s Hz based on available SLMs and stimulation parameters. Finally, to yet further expand the addressable population, we commissioned and directed the development of high-speed, high-pixel-count SLMs for precise, large-scale 3D two photon optogenetics (supported under our DARPA Neuro-FAST program from Dec. 2013 onward). Applying our custom SLMs, along with low magnification objectives (e.g., 10x 0.6NA), we discovered that we can simultaneously photo-stimulate neurons across >1mm of cortex, an unprecedented range with substantial implications. We demonstrate and validate these technologies in awake mouse cortex, wherein the same neurons are imaged and stimulated across weeks. Neurons that were imaged and stimulated *in vivo* were later found in registered CLARITY-processed tissue, thus integrating anatomical characterization (of major significance for cell type specific labeling). Together, these novel approaches enable all-optical cellular-resolution read/write, activity-guided, and potentially closed-loop control of precisely emulated activity patterns (coupled with molecular and anatomical information from the same volume), which will be of critical significance for understanding how behavior arises from circuit activity patterns.

Disclosures: J.H. Marshel: None. **S. Quirin:** None. **E.L. Sylwestrak:** None. **A. Chibukhchyan:** None. **A. Crow:** None. **C. Ramakrishnan:** None. **K. Deisseroth:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.17/WW46

Topic: I.04. Physiological Methods

Support: HHMI

NIH

DARPA

Simons Foundation

Title: Two-photon calcium imaging in behaving rhesus macaque and its feasibility for all-optical brain-machine interfaces

Authors: *X. SUN, E. TRAUTMANN, D. O' SHEA, J. MARSHEL, W. ALLEN, I. KAUVAR, C. RAMAKRISHNAN, S. RYU, K. DEISSEROTH, K. SHENOY
Stanford Univ., Stanford, CA

Abstract: Optical techniques such as two-photon (2p) calcium imaging have the potential to transform the way we interrogate neural circuits, both in the realm of basic neuroscience and in the development of brain-machine interfaces (BMIs) by overcoming some of the limitations of electrophysiological methods. We have developed a platform for performing long-term, stable 2p imaging in behaving rhesus monkeys during a motor task.

There are many challenges to translating 2p imaging from rodents to monkeys. To screen for GCaMP constructs that express at appropriate levels in monkeys, we injected three constructs of different volumes at 6 separate sites in PMd and M1. We monitored GCaMP expression after virus injection with wide-field and 2p imaging. To maintain the optical clarity of the imaging chamber, we developed a chamber that incorporates a transparent replaceable silicone window sealed from the external environment to minimize risks of infection. We minimized the chamber size to support bone health and surgical feasibility, without compromising accessible imaging area using large multiphoton lenses with short working distances. Finally, we developed a novel rigid three-point head restraint system that prevents the task itself from generating motion artifact and a brain stabilization system that applies gentle pressure to restrict total motion throughout multi-hour behavioral experiments. Using an image of surface vasculature acquired by wide-field imaging as a map, it is possible to reliably navigate to specific cortical locations using blood vessels as landmarks. As a result, we are able to visit each injection site repeatedly in a targeted way across imaging sessions.

While systems for 2p imaging in behaving monkeys are progressing, assessing their potential for all-optical BMIs is of interest. A quantitative comparison between imaging and electrode data ideally from the same animal, as well as their BMI decoding performance, has yet to be performed. As a first step, we compared these modalities by decoding hand trajectories using Kalman-filter based decoders from synthetic 2p calcium imaging signals generated from Utah array recordings. We evaluated how realistic, but simulated, imaging signals from hundreds of neurons encode kinematic information in comparison with the measured action potentials. We found that it was possible to decode hand position and velocity from synthetic imaging signals, where decode quality varied considerably as a function of SNR and the imaging frame rate. This simulation provides insights into the value of applying optical techniques to NHP studies, and should help establish the feasibility and design space for all-optical BMIs.

Disclosures: **X. Sun:** None. **E. Trautmann:** None. **D. O' Shea:** None. **J. Marshel:** None. **W. Allen:** None. **I. Kauvar:** None. **C. Ramakrishnan:** None. **S. Ryu:** None. **K. Deisseroth:** None. **K. Shenoy:** F. Consulting Fees (e.g., advisory boards); Neuralink Inc., consultant, Cognescent, Scientific Advisory Board, Heal, Scientific Advisory Board.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.18/WW47

Topic: I.04. Physiological Methods

Support: NINDS F32NS095690-01

NINDS DP2NS087725-01

Title: Precise spatiotemporal control of neural activity

Authors: ***A. R. MARDINLY**[1], I. OLDENBURG[2], N. PEGARD[2], S. SRIDHARAN[2], E. LYALL[2], K. CHESNOV[2], S. BROHAWN[2], L. WALLER[2], H. ADESNIK[2]
[1]UC Berkeley, El Cerrito, CA; [2]Univ. of California, Berkeley, CA

Abstract: Neural circuits encode information in the rate, timing, and number of action potentials and in the identity of active neurons. A technological approach that could drive precise spatiotemporal trains of neural activity with cellular resolution and millisecond temporal precision in full 3D could unlock a new class of experiments to probe the coding logic of neural circuits. Recently developed all-optical multiphoton approaches combine spiral scanning on C1V1-expressing neurons with 2P calcium imaging to probe the function of neural circuits. While these approaches effectively active specific groups of neurons with high spatial resolution, they do not possess the requisite temporal precision to drive complex, naturalistic sequences of spatiotemporal neural activity. To overcome this challenge, we developed new opsins for scanless 2P stimulation and combined them with a novel form of 3D scanless temporal focusing called 3D-SHOT. We introduce ChroME, a new ultra-fast opsin that is 3.5-fold more powerful than Chronos. Brief (<5 ms) 3D-SHOT stimulation of ChroME-expressing L2/3 pyramidal neurons generates reliable action potentials with sub-millisecond jitter. Using *in vivo* cell attached patch recordings, we demonstrate high fidelity replay of complex, naturalistic sequences of spatiotemporal neural activity in pyramidal neurons using ChroME and in PV, SOM, and VIP interneurons using Chronos. We combine 3D holographic stimulation with volumetric calcium imaging to probe the network response to introducing varying numbers of action potentials into the cortex while holding stimulation rate constant. Finally, we validate that spatial resolution is maintained while volumetrically stimulating up to 50 spots simultaneously, and all-optically activate groups of neurons with millisecond precision. By combining new ultra-fast, powerful

opsins with novel scanless 3D stimulation paradigms, we gained temporal control of multi-photon all-optical experiments, paving the way to systematically activate functionally defined neurons with parameterzed stimulus patterns during simultaneous calcium imaging in the awake, behaving brain.

Disclosures: A.R. Mardinly: None. **I. Oldenburg:** None. **N. Pegard:** None. **S. Sridharan:** None. **E. Lyall:** None. **K. Chesnov:** None. **S. Brohawn:** None. **L. Waller:** None. **H. Adesnik:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.19/WW48

Topic: I.04. Physiological Methods

Title: Automated fine-scale three-dimensional paw tracking and posture classification system in mice

Authors: *M. BALBI, A. LUO, L. BOLANOS, F. BOLANOS, J. LEDUE, T. H. MURPHY
Psychiatry, Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Forelimb movements represent a refined feature of the mammalian motor system. Goal-directed limb movements integrate relevant sensory inputs and motor commands. The resulting movement trajectory and kinematics lie in a high dimensional space, which is intractable for manual rating. We developed an integrated hardware and software system that uses low-cost, single-board computers, machine vision, and supervised learning to automate paw tracking and posture classification. Head-fixed mice were trained to pull a lever for a water reward upon hearing an auditory ¨go¨ cue while being recorded by a two-camera stereovision system. Previous implementations of machine vision for paw movement analysis only track the centroid of the paw, as the high deformability of the paw imposes a great challenge on automatically discriminating posture. To extract posture information from videos, we developed machine vision techniques to track a fluorescently dyed (Nile Blu 1mg/ml, emission: RG9 -IR-) mouse paw from the videos, extract its geometric and kinematic features, and reconstruct the 3D movement trajectory during contact, grasping and release of the lever. Based on preliminary data, the range of mouse paw postures was clustered into 9 different types, characterized by their static (relaxed-on, contracted-on and off lever) and dynamic features (along the 3 spatial dimensions). We trained nonlinear classifiers (random forests and neural networks) using these features and 3D trajectories to estimate paw posture parameterized with openness, digit extent, and contact with the lever. In our study, we validated the robustness and precision of our system against a physical readout of lever position. Our approach allows for longitudinal automated assessment and comparison of fine-scale paw movements in health and disease.

Disclosures: **M. Balbi:** None. **A. Luo:** None. **L. Bolanos:** None. **F. Bolanos:** None. **J. Ledue:** None. **T.H. Murphy:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.20/WW49

Topic: I.04. Physiological Methods

Support: CIHR MOP-12675

CIHR FDN-143209

Title: Correlating mesoscopic cortical calcium activity with single unit activity or body movements in mice

Authors: ***M. P. VANNI**[1], D. XIAO[1], C. MITELUT[1], A. W. CHAN[1], J. LEDUE[1], J. BOYD[1], M. BALBI[1], F. BOLANOS[1], Y. SEKINO[1], G. SILASI[1], N. V. SWINDALE[2], T. H. MURPHY[1]
[1]Dept. of Psychiatry, Univ. of British Columbia, Vancouver, BC, Canada; [2]Univ. British Columbia, Vancouver, BC, Canada

Abstract: Resting state brain spontaneous activity is commonly used to map the brain at mesoscale in a large number of species. With the recent development of mice expressing new genetically encoded calcium indicators (GCaMP6), cortical activity can be measured at high sensitivity in awake behaving animal at low cost in contrast to most other approaches. A new challenge is now the possibility to combine it with multiple parallel measurements ranging different spatial and temporal scales from single neuron electrophysiology to behavior. Here, we explored the arrangement of mesoscale spontaneous calcium activity in cortex firstly with single cell activity and secondly, with behavior. Mice expressing the calcium indicator GCaMP6 were implanted with bilateral chronic window covering most of the dorsal cortex including motor, somatosensory and visual cortex as well as medial region such as retrosplenial and cingulate cortex. Green fluorescence under blue LED illumination was recorded in awake mice with a CCD camera. To remove the contribution of blood artefact, green or blue reflectance was also recorded in parallel to estimate changes in cerebral blood volume (CBV). Single unit electrophysiology was recorded using chronically implanted tetrodes or laminar 64channels electrodes in different cortical and subcortical regions of the brain. Behavior measurement was performed using video analysis of the paws, whiskers or pupil movement. The cortical sequences of activity associated with each individual neuron was mapped by averaging the fluorescence fluctuation preceding and following each individual spike. We observed that, in contrast to cortex, subcortical neurons exhibit more complex cortical motifs of activity with the presence of strong suppression. By analyzing the cortical dynamic in different cortical regions, motif

classification could be performed. Similarly to spike triggered averaging, mapping could also be performed by using correlation between firing rate and calcium activity. Time jittering of these signals provides information about the temporal dynamics of each signal. Based on this strategy other modalities were combined with mesoscopic imaging such as motor action. By quantifying the density of movement of forepaw or whiskers, behavior triggered averaging or multimodal correlation showed a precise mapping involving forelimb or barrel somatosensorimotor cortex respectively. This multimodal, multiscale approach could be used to explore functional remapping in multiple models of disease such as ischemia. In future, this could be extended to other modalities such as peripheral nerve recordings or mouse vocalization.

Disclosures: **M.P. Vanni:** None. **D. Xiao:** None. **C. Mitelut:** None. **A.W. Chan:** None. **J. Ledue:** None. **J. Boyd:** None. **M. Balbi:** None. **F. Bolanos:** None. **Y. Sekino:** None. **G. Silasi:** None. **N.V. Swindale:** None. **T.H. Murphy:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.21/WW50

Topic: I.04. Physiological Methods

Support: CIHR FDN-143209

CIHR CGSM

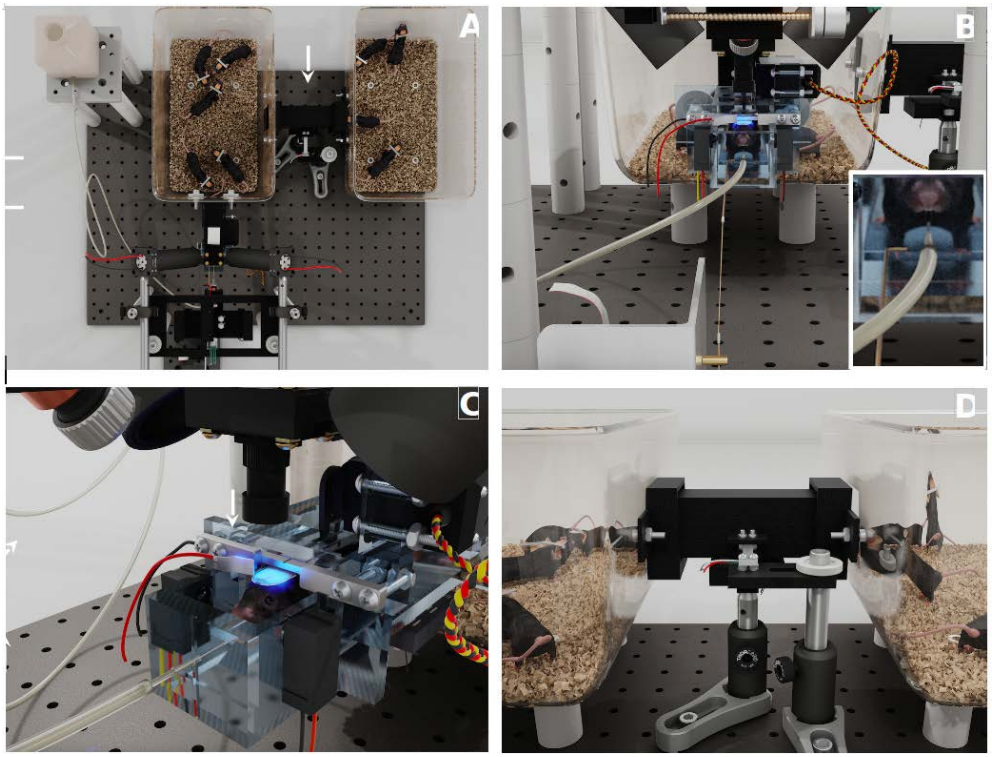
Brain Canada Neurophotonics Platform

Title: Automated optogenetic and mesoscopic brain imaging system for the mouse home-cage with web-based monitoring for up to 10 mice

Authors: *F. BOLANOS, J. M. LEDUE, J. D. BOYD, T. H. MURPHY
Psychiatry, UBC, Vancouver, BC, Canada

Abstract: Over the last several years experiments that rely on awake and behaving mice have become a new standard. These experiments can require the mouse to be head-fixed to obtain high quality optical data and are usually done chronically following many mice over the course of several weeks which can induce stress. Additionally these mice have to be trained to tolerate long sessions of head restraint, and since this is done for each mouse it can become a time consuming task for the experimenter. Here we describe a home-cage based system that automatically identifies, head-fixes, rewards and weighs each mouse. The system is similar to the automated imaging system that we published before (Murphy et al. 2016), but differs in its head-fixing mechanism and in the addition of a movable laser to stimulate different areas of the cortex while the mouse is head-fixed and involves to 2 linked cages increasing mouse number to

potentially 10. We followed 7 mice for 2+ months, 6 of the 7 mice learned to auto head-fix within a week. We obtain an average of 20 minutes of head-fixed imaging data per day, per mouse yielding a total of 2 hours of imaging data per day/cage. Additionally, we obtain ~10 weight data points per day per mouse. The system can support up to ten mice that are automatically imaged, stimulated and weighed. We also describe a training protocol where the RFID identified mice learn to self-initiate brain imaging trials in order to obtain water rewards while their movement becomes progressively more restricted until they are fully head-fixed. The system utilizes the Raspberry Pi single board computer in order to minimize cost and thus maximize the potential to scale up the system (see rendering).



Disclosures: **F. Bolanos:** None. **J.M. LeDue:** None. **J.D. Boyd:** None. **T.H. Murphy:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.22/WW51

Topic: I.04. Physiological Methods

Support: MOP-12675

FDN-143209

Title: Sub-cortical or peripheral nerve spike-triggered cortical mesoscale activity associated with specific actions in awake chronic mice

Authors: *D. XIAO[1,2], J. M. LEDUE[1,2], J. GREWAL[3], T. H. MURPHY[1,2]
[1]Kinsmen Lab, Dept. of Psychiatry, Vancouver, BC, Canada; [2]Djavad Mowafaghian Ctr. for Brain Health, Univ. of British Columbia, Vancouver, BC, Canada; [3]Dept. of Psychiatry and Brain Res. Ctr., Vancouver, BC, Canada

Abstract: Flexible control of voluntary movement relies on hierarchical cortical and subcortical neuronal circuits to mediate sensorimotor and memory information integration. How information processing in the motor pathways eventually drive a specific action still largely unknown. The difficulty comes from recording large scale neural activity in awake mice and at the same time as accurately monitoring the motor output. Here we provide a new design of chronic recording system which integrates with wide-field GCaMP imaging, multi-site sub-cortical/cortical cellular electrophysiology and peripheral nerve recording in head-fixed free running mice. Facial motor nerve impulse was measured by paired fine wire (15 μm) recording directly from the nerve dominating the whisker. Multiple tetrodes were implanted in region of interests to record extracellular activity in motor pathway (can be thalamus, striatum or brainstem). A mesoscale nerve spike-triggered averaging procedure allowed the identification of motifs that are preferentially related to motor output by employing genetically targeted indicators of neuronal activity. Preliminary results indicated self-initiated vibrissa movement not only driven by primary sensorimotor areas (barrel cortex, motor cortex) but also involved in association cortical regions (retrosplenial cortex, secondary motor cortex) and striatum. Motor feedback in the cortex can also be investigated by microstimulation in the motor nerve through the same recording electrode. Our chronic recordings remained stable for weeks, demonstrating that this method can be employed to investigate the dynamic and distributed neuronal ensemble interactions that underlie processes of motor control and sensorimotor learning in behaving mice.

Disclosures: D. Xiao: None. **J.M. LeDue:** None. **J. Grewal:** None. **T.H. Murphy:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.23/WW52

Topic: I.04. Physiological Methods

Support: Canadian Institutes of Health Research (CIHR) Operating Grant MOP-12675

Canadian Institutes of Health Research (CIHR) Foundation Grant FDN-143209

Title: Movement initiation in gcamp6 mice is preceded by stereotyped, multi-second wide-scale dorsal cortex dynamics

Authors: *C. MITELUT[1], A. X. LUO[1], G. SILASI[2], Y. SEKINO[3], J. BOYD[1], F. BOLANOS[1], N. V. SWINDALE[1], T. H. MURPHY[1]
[1]Univ. of British Columbia, Vancouver, BC, Canada; [2]Univ. of Ottawa, Ottawa, ON, Canada; [3]Grad. Sch. of Med. Sci., Kyushu Univ., Fukuoka, Japan

Abstract: Modern experimental neuroscience has sought to identify the neural correlates of free, or voluntary, behaviour largely focusing on instructed task paradigms in humans using fMRI or EEG (Haggard 2008).
Using GCaMP6 transgenic mice (Ai93 and Ai94) we sought to identify neuronal dynamics preceding the initiation of voluntary behaviours (i.e. lever pulling, grooming and licking) by recording calcium activity in dorsal cortex along with simultaneous recordings of a lever deflection angle in a pulling task (11/11 mice) and behavioral videos (5/11 mice). To automate detection of grooming and licking behaviours using video annotation, we segmented the foreground from each frame, projected it to PCA space and used binary Random Forest classifiers to detect behaviour initiation. During each ~20 minute recording session (30-50 sessions/mouse) we identified quiescent periods (i.e. animal was motionless for > 3s) preceding lever pulling (~20-100/session), grooming (10-40/session) and licking (10-30/session) and evaluated calcium activity (dF/F0) in three cortical ROIs: motor, limb and retrosplenial (RS). We found that lever pulling was preceded by limb cortex calcium activation of up to 2% as early as 7 seconds (or more) prior to movement initiation - consistent with findings of early prediction of cognitive performance in humans (Soon 2008). In contrast, grooming was preceded by ~1s of cortical activation and licking by 2-3s. Interestingly, rather than arising from a ~0% dF/F0 baseline and following monotonic activation of limb and motor cortex - all behaviours were initiated during the second activation phase of an activation-depression-activation cycle in limb cortex. This suggests that spontaneous behaviours arise from ongoing large-scale oscillations.
We additionally found that during lever pulling cortical activation peaked at different times in different ROIs: RS activation peaked 1sec before behaviour initiation, whereas motor cortex activation peaked on behaviour initiation (i.e. t=0s) and limb cortex 1s after initiation. This suggests that the distinct roles cortical regions play in complex behaviour preparation and execution can be studied at the mesoscale level using GCaMP6 signals.
These results are consistent with research in humans that behaviour preparation begins substantially earlier than is available to higher order faculties (e.g. awareness in humans). The findings further validate the use of widefield imaging in GCaMP6 mice to study learning and behaviours during task acquisition paradigms and the further investigation of spontaneous behaviour in non-human animals using task-free, non-reporting paradigms.

Disclosures: C. Mitelut: None. **A.X. Luo:** None. **G. Silasi:** None. **Y. Sekino:** None. **J. Boyd:** None. **F. Bolanos:** None. **N.V. Swindale:** None. **T.H. Murphy:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.24/DP15/WW53 (Dynamic Poster)

Topic: I.04. Physiological Methods

Support: NSF EAGER IOS-1611090

Title: An active nanophotonic multi-beam probe for optogenetic stimulation *In vivo*

Authors: *A. KEPECS[1], Q. LI[1], A. MOHANTY[2], A. M. TAYADON[2], M. LIPSON[2]
[1]Cold Spring Harbor Lab., Cold Spring Harbor, NY; [2]Columbia Univ., New York, NY

Abstract: The ability to optically turn on or off the activity of specific neural populations has revolutionized the investigation of neural circuits. Most applications, however, employ a single optical fiber to flood a large area of the brain with light, limiting the ability to selectively activate neurons with high spatial resolution in vivo. Using silicon photonics technology it is possible to miniaturize complex optical circuits on a silicon chip, resulting in a quantum leap in optical control, matching the scale and resolution currently achieved by electrical silicon probes for electrical recordings.Here we developed and tested the first visible light multipoint-emitting nanophotonic switches for in vivo optogenetics. To enable selective activation of neurons using multiple, independently controllable optical beams, we used micro-switches that could route light toward different diffraction gratings. Fast and independent control over each beam was achieved via external micro-controllers. We demonstrated that the same technology previously used for switching light in the near infrared spectral range could be used for switching light in the visible, using the thermo-optic effect of silicon nitride, a material that is transparent to the visible spectral range.To characterize the probe performance in mammalian brain in vivo, anaesthetized mice expressing ChR2 in PV+ inhibitory interneurons (PV-Cre x Ai32 mice) is used. The demonstrated probe employs a 1x8 multipoint-emitting nanophotonic switch (emitter size: 20 x 20 µm, 250 µm interbeam distance) coupled to a tungsten electrode array. The probe could be readily inserted and lowered into cortex. By applying two external microcontrollers to arbitrarily direct 472nm blue light to 4 output grating light emitters we could independently activate single ChR2-expressing PV interneurons throughout layers 2-6 of visual cortex in vivo. The activated PV interneurons showed robust spike firing with short first spike latency (1.475 +/- 0.063 ms, mean +/- sem) and low jitter (0.031 +/- 0.003 ms, mean +/- sem). In conclusion we successfully show our ability to use external microcontrollers to arbitrarily direct light to multiple densely spaced illumination points along the shank. Scaling it up to a larger number of beams (e.g.128) is expected to be straightforward since these devices are defined lithographically, enabling high-resolution optical stimulation in deep brain regions.

Disclosures: **A. Kepecs:** None. **Q. Li:** None. **A. Mohanty:** None. **A.M. Tayadon:** None. **M. Lipson:** None.

Poster

437. Optical Methods for Functional Circuit Analysis *In Vivo*

Location: Halls A-C

Time: Monday, November 13, 2017, 1:00 PM - 5:00 PM

Program#/Poster#: 437.25/WW54

Topic: I.04. Physiological Methods

Support: NIH Grant U01MH109091

Title: Transgenic sparse expression of genetically

Authors: ***S. D. ANTIC**[1], C. SONG[2], M. COLAVITA[2], T. KNOPFEL[2]
[1]Neurosci, UConn Hlth., Farmington, CT; [2]Med., Imperial Col. London, London, United Kingdom

Abstract: One of the major goals of the BRAIN Initiative is development of the tools for recording electrical activity in many neurons simultaneously. Genetically encoded optical indicators, particularly genetically encoded voltage indicators (GEVIs), enable longitudinally monitoring neuronal circuit dynamics of identified cell populations, and advanced transgenic approaches achieve high levels of indicator expression in vivo. GEVI's membrane delimited signals arise largely from the neuropil where dendritic and axonal membranes of many cells intermingle, hence targeting non-sparse cell populations leads to dense expression patterns such that optical signals from neuronal processes cannot be allocated to individual neurons. Here we show that sparse but strong expression of genetically encoded optical indicators, using a titratable recombination-activated transgene transcription, achieves a Golgi staining-type indicator expression in vivo, and such expression pattern is capable of further resolving morphologies of individual cells including dendritic spines and axonal boutons. Using different transgenic strategies, we also illustrate that co-expression of genetically encoded voltage and calcium indicators can be achieved in the same animal for studying neuronal circuit input-output relationships. These transgenic strategies serve as ideal approaches for GEVI-based optical voltage monitoring of neural circuits at the level of groups of neurons or at the level of single neurons in intact mammalian tissues. Equally important, the sparse GEVI technology allows monitoring voltage transients of specified cell types such as pyramidal neurons in a specific cortical layer, thus further refining the experimental tools for dissection of brain circuits.

Disclosures: **S.D. Antic:** None. **C. Song:** None. **M. Colavita:** None. **T. Knopfel:** None.